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## FASCIOLOPSIS BUSKI

A PARASITE OF MAN AS SEEN IN SHAOHING, CHINA \*

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### THE DISEASE

Fasciolopsis infestation of man, for many years a medical curiosity, has within the last decade come to be recognized as a serious condition both for the individual and for the community concerned. The first flukes of this type to be described were discovered by Busk in the intestines of a Lascar sailor dying in England in 1843. The parasite has been found in a few other cases in Europe and America, but apparently the disease is endemic only in tropical and subtropical countries. As these comparatively little known regions become more deeply penetrated by the medical missionary and others, infestation of man by this species or related ones is found with increasing frequency, and is now known to occur in India, Assam, Siam, Natal, Borneo, Straits Settlements, Sumatra, Cochin-China, Tonkin, and along the coast of China (Canton, Hongkong, and as far north as the Yang-tse Valley, where in the Shaohing district it is particularly prevalent). In pigs *Fasciolopsis buski* has been reported to be very common in Hongkong and Tonkin. In man, at least in Shaohing, the disease appears to be more common in early life, 5 to 20 years, but it is not rare in persons up to middle life, and we have found it in one infant but a year old.

The prevalence of this disease in any community will in all probability depend on the dietetic habits of the people. Dobson reports that 1 per cent. of the stools of 1,000 coolies examined in India showed the eggs present; Mathis and Léger speak of the "extreme rarity" of the infection in man in Cochin-China. In Shaohing, China, it is extremely common. In the seventeen months from January, 1908, to May, 1909, the diagnosis of fasciolopsis infection was confirmed by microscopic examination of the feces in  $5\frac{1}{2}$  per cent. of all the dispensary patients, about 2 per cent. more presented very suggestive symptoms, and doubtless many others were harboring the parasite, though without symptoms. Shortly after the in-patient department of the hospital was

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opened nineteen out of twenty patients, or 95 per cent., showed ova in the feces, though none of the twenty was admitted for that condition, nor even suspected it. During the years 1914-16 out of 304 cases admitted to the Christian Hospital, and in which routine fecal examinations were made, 87, or 28 per cent., were found positive for the presence of fasciolopsis.

Three stages may be recognized clinically.

1. *The Period of Latency.*—This period is without notable symptoms and may occupy months or even years. A few flukes in the intestine seem to cause no inconvenience, and how severe the infection needs to be before giving rise to symptoms it would be impossible to say; but in the Christian Hospital the number of flukes recovered from the stools of an individual has varied from a few tens to over three thousand (3,328), and while in general the severity of symptoms is proportional to the number of flukes present, there is also a marked difference in the degree of resistance that an individual may possess, some from whom but a few hundred were recovered being clinically in worse condition than those who had harbored as many thousands. It is probable that asthenia in varying degree, and a mild anemia may appear toward the termination of this stage.

2. *The Period of Diarrhea.*—During the second stage diarrhea appears, and is the condition for which most often relief is sought. There is generally a history of five or six stools a day extending over a period of months, and often with intermissions of days or weeks during which the bowels act normally. The stool itself is usually light yellow in color, without any evidence of blood even under the microscope in uncomplicated cases. It contains a considerable amount of undigested matter, and has a peculiarly offensive odor. Anemia now becomes noticeable and may be extreme, and in Shaohing the combination of anemia with chronic diarrhea is practically pathognomonic of fasciolopsis infection. Other symptoms are inconstant. The appetite may be impaired or even increased, but is usually unaffected; occasionally there may be dull aching pain distributed throughout the abdomen or localized in the duodenal region; temperature, pulse, and respiration do not in uncomplicated cases differ from the normal. In infants and young children the abdomen is very protuberant, and may be the first evidence of disease noted by the parents.

3. *The Period of Edema.*—During the third and final stage anemia is always marked but the most prominent as well as most distressing symptom is edema. This usually affects first the abdominal cavity, then extends to the genitals (which may be very greatly distended), and to the lower extremities beginning in the feet and ankles but soon involving the whole limb, and finally appears in the upper extremities,



the face and the lungs, and with these advanced conditions insufficiency of the cardiac valves may supervene. To relieve the dyspnea due to the ascites it may be necessary to resort frequently to paracentesis abdominis, sometimes as often as every five or six days for a considerable period. The urine is normal except for undue concentration, and even under diuretics may be reduced to but two or three ounces in twenty-four hours. The skin has a yellowish tinge, is harsh and dry, the tongue is glazed, the temperature has a tendency to fall one or two degrees below normal, the patient become extremely weak and death when it occurs is apparently due to exhaustion.

In the treatment of this condition turpentine, oil of eucalyptus with chloroform, thymol and beta-naphthol have all been used with success, the treatment being in fact the same as for hookworm. Personally, I favor beta-naphthol, and believe it better practice, on account of the marked depression which occasionally comes on even when least expected, whatever drug is employed, to give small doses repeated as often as necessary, rather than to attempt to expel all the parasites in one or two doses. Oil of chenopodium has not yet been tried, because it was unavailable when called for. Restricted diet and saline purgatives both before and after are of course indicated. Dead flukes will usually begin to be passed within twelve hours after taking the anthelmintic, and will continue to come for two or three days. Tonics are rarely needed, as it is astonishing how rapidly the anemia disappears once the intestines are cleared.

*Prophylaxis*, naturally, will consist in the avoidance of uncooked food. The life history of this fluke is still to be worked out, but the fact that fresh water snails enter largely into the diet of the people of this region, and that before being eaten they are subjected to only slight scalding, is suggestive that these molluscs may be the secondary host. Shrimps have also been considered, but they are in general quite thoroughly cooked.

#### THE PARASITE

The parasite in question exhibits such wide variations in morphology that four species have been described, and recently Brown has suggested a reclassification into two groups differentiated chiefly by the presence or absence of cuticular spines. The literature on the subject is somewhat contradictory and confusing, and until recently the material has been so scanty and appeared at such great intervals of space and time, that a restudy of the subject in the light of considerable clinical experience and with a larger amount of laboratory material seemed likely to yield important results. The following report, which is preliminary to further studies on the life history of the parasite and its effects on man from the pathological standpoint, is based on a

practice extending over several years in a heavily infested district (Shaohing, China), supplemented by a laboratory study of 433 specimens. Twenty-one flukes have been cleared, the ventral sucker with cirrus sac and metratrum attached have been dissected out from three, ova have been removed for measurement from the lower uterus of nine, and seventeen series of microscopic sections have been made of individuals conforming to the descriptions of the three types which have been described as *F. buski*, *F. rathouisi* and *F. goddardi*, including five serial sections in different planes.

In selecting specimens as representatives of each type, in general, those were chosen which in size fell within the published measurements; viz. a length of 25 mm. or more for *F. buski*; 21-24 mm. for *F. goddardi*, and 15-20 mm. for *F. rathouisi*. But a few exceptions were made where other physical features clearly indicated a different classification. This fact will explain the apparent inconsistencies in the grouping, and illustrate at the same time the extreme difficulty encountered in making any distinctions on account of the many borderline cases.

In summarizing the results of this study, it will be convenient to consider first the variations upon which differentiation into species has been based; and to conclude with a somewhat detailed description of the morphology as a whole, especially of those features about which there has hitherto been doubt.

The variations which have figured in the differentiation into separate species may be grouped as follows:

1. *Color and Consistency*.—Specimens preserved in alcohol vary in color from brown to grayish-white, and frequently the vitellaria are clearly outlined by a bluish black pigmentation along the lateral and posterior margins (Pl. XII, A). Some are firm in consistency, while others are flabby and soft. These variations are found in individuals of all the types of this genus which I have examined, and are undoubtedly due to postmortem changes. The fluke when alive or freshly killed is of a deep pink color, not unlike that of boiled ham, and the great majority of all flukes recovered from the stools are of this color and are firm in consistency. Occasionally flukes are passed which are pearly white with dark borders, and are flabby in appearance, and to the touch. Under the microscope such specimens are seen to have lost their cuticula (Pl. XIV, A), and the cells of the yolk glands have fallen away from the basement membrane of the acini and are collected in a more or less disintegrated mass in the lumen, the nuclei being deeply pigmented. Recalling the fact that flukes are often two or three days in the intestine after the anthelmintic has been taken, it is easy to understand that partial digestion of the surface has occurred, quite sufficient to account for the phenomena observed. Similar but



less marked changes occur in specimens allowed to remain for a considerable time in water before being placed in preservative.

2. *Size and Shape*.—Variations in size and shape are so extreme (Pl. XII, *A*) as to warrant a belief in the existence of more than one species until it is found that gradations from one to another type are so gradual as to make lines of demarcation quite impossible.

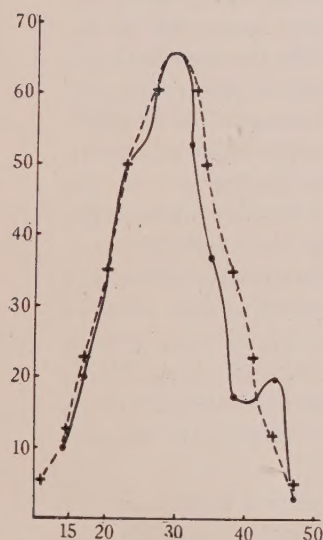


Fig. 1. Length frequency curve constructed from measurements of 378 individual flukes.

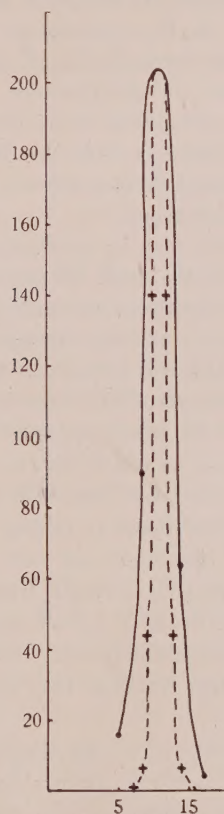


Fig. 2. Width frequency curve.

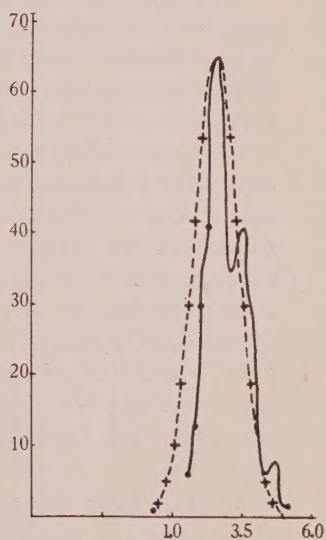


Fig. 3. Proportion frequency curve, i. e., ratio of length to width.

In this investigation measurements were made of 378 flukes ranging from 13 to 48 mm. in length and from 4 to 17 mm. in width. Progression in length and width measured in millimeters, and in the ratios between them, measured in  $\frac{5}{100}$  of the width was found to be perfectly even. Frequency curves were then plotted and are shown in Graphs 1, 2 and 3. A certain amount of error is possible here, due to the fact that the flukes were not all in the same state of muscular contraction when fixed, but all the curves, for length, for width, and

for proportion, are so distinctly unimodal, and conform so closely to the corresponding theoretical curves for variations\* as to leave little doubt that we are dealing with a single species.

The thickness measured at the middle varies in this series from 0.8 to 3.0 mm., while over the acetabulum, the thickest part, the maximum found was 3.5 mm., and the variation is somewhat less. No relation can be found between the thickness and the various suggested species. That described as *F. rathouisi* is characteristically "short and stocky"—one such measured 20x12x2.8, was brown in color, firm in texture, and in a state of opisthotonos. But another measures 19x13x2.0, is glistening white with dark borders, and lies flat and relaxed. On the other hand, a fluke (*e* in Pl. I, *A*) typical of *F. buski*, measured 43x12x1.0, but others measuring 40x9x2.3 or 32x10x2.3 or 33x10x3.0 also occur.

In part, these differences may be ascribed to natural variation, but some are certainly due to the state of muscular contraction at the time of fixation. This is apparent not only from the numerous and deep transverse rugae which are seen frequently in preserved specimens (cf. *i* and *j* in Pl. XII, *A*), but also from observations on flukes in the fresh state. In one instance 22 flukes were brought in immediately after evacuation, all of which appeared to be long and narrow (about 35x8 mm.) and many of which showed an inrolling of the edges of the cephalic portion about the long axis so as to form almost a complete sheath. I was interrupted in taking their measurements, and after remaining in water four hours all were found to have assumed the ordinary appearance, varying in length from 18 to 35 mm., in width from 13 to 20, the average being 28.6x16.4, and extremes being 35x20, 18x16 and 28x13. On another occasion a fluke measuring 28x19 on evacuation, was found after standing two days in water to measure 41x17.

3. *Head Cone*.—In none of the Shaohing flukes is there a distinct "head cone" or shoulder such as occurs in *Fasciola hepatica*, for example; but in certain specimens (cf. *i*, *g*, *f* in Pl. XII, *A*) there is a narrowing at the level of the genital pore, and when viewed from the side a posterior bulging. This is due to the contraction of the dermo-muscular tube and parenchymal muscles closely about the solid muscular bodies of the region (oral sucker, pharynx and acetabulum), and is not distinctive of any of the three types.

\* The normal curve in all these figures is plotted according to the commonly used formula

$$y = \frac{n}{\sigma \sqrt{2\pi}} \cdot \frac{1}{e^{\frac{x^2}{2\sigma^2}}}$$



4. *The Cuticula*.—In the early description of *F. buski* by Cobbold, the integument is said to be "smooth and unarmed," and Odhner, Ward, Looss, Braun and Rodenwaldt either state that spines are absent or adopt the view generally held that they are absent. Heanley and Jefferys state positively that they are present "though very difficult to find in some mounted specimens" (Heanley), and Leiper says they are present in the species *F. rathouisi* and *goddardi*, and explains their occasional absence from certain specimens by the "deciduous character" of the cuticula, claiming to have seen in a section of Rodenwaldt's specimen the regularly recurring pits in the cuticula from which the spines have dropped out. More recently Brown has found an apparent coincidence between the presence or absence of spines and the marked differences in gross appearance already discussed, and suggests a reclassification of the several varieties now named into two groups on the basis of this characteristic.

Of the present series of 433 flukes, half (including all the variant types illustrated in Pl. XII, *A*, and among them typical examples of the two groups suggested by Brown), were examined by strong reflected and transmitted light, and in all of these spines were found though in many cases only with difficulty and in very small numbers, due to the deciduous character of the cuticula to which Leiper has already called attention.

Of the flukes photographed in Plate XII, *A*, *e*, *f*, *j* and *k* have since been imbedded and sectioned. In *j* and *k* the cuticle is well preserved and the spines numerous; in *e* and *f*, on the contrary, the cuticle, except in a very few scattered spots, is entirely gone. A comparison of Plate XII, *B*, Plate XIII and Plate XVII, *A*, shows how readily even in fairly well preserved specimens the cuticula strips off from the basement membrane carrying the spines with it. These facts furnish a satisfactory explanation of the occasional failures hitherto to demonstrate the presence of cuticular spines, and warrant the conclusion that they are charactersitic of the entire genus.

5. *Form and Size of Various Viscera*.—The cirrus sac is convoluted in all specimens, varying from two or three close turns like the strands of a rope to larger spirals, and its course may be straight or sharply bent upon itself; but neither these variations nor the relative length of the sac posterior to the acetabulum are characteristic of any group, being found in each. The diverticulum of the seminal vesicle, the so-called "cecal appendage" was present in all specimens examined.

The shellgland is an elastic body, and though normally ovoid in shape, is capable of being markedly affected by the surrounding muscles; appearing in one highly contracted specimen (fluke *m*) as an antero-posteriorly flattened disk extending the entire dorsoventral diameter of the body.

TABLE 1.—VISCERAL MEASUREMENTS IN MICRONS. LETTERED INDIVIDUALS REPRESENT SECTIONS; THE OTHERS CLEARED FLUKES

Flukes: Cleared and Sectioned	Oral Sucker		Pharynx	Genital Pore	Ventral Sucker			Shell Gland			Cirrus Sac*		Testes	Vitel- line Acini	Remarks
	Whole Organ	Orif- ice			Whole Organ	Orifice	Dis- tance from Head	Size	Dis- tance from Head	Dis- tance from Head	In Microns	Per Cent.			
F. buscki	45 × 13 × 1.3	350	759 × 992	.....	3,212 × 2,599	876 × 1,518	1,460	1,401 × 1,576	6,494	15 mm.	8,796	50	292-759	58-175	
	33 × 8	175	467 × 700	116 × 292	2,511 × 2,102	1,051 × 1,216	1,732	992 × 1,188	7,884	12 mm.	6,672	59	? - 492	58-146	
	29 × 11	280	408 × 759	234 × 408	2,920 × ?	876 × 1,460	1,284	1,109 × 1,226	7,416	12 mm.	4,204	54	116-700	58-116	Cirrus sac very angular course
	97 × 9	..	..	175 × 408	2,326 × ?	467 × 584	1,284	934 × 1,168	6,716	12 mm.	8,212	47	116-525	58-175	
	29 × 13	408	584 × 700	..	2,698 × 2,336	..	1,576	..	..	..	..	..	..	..	
	32 × 11	350	642 × 759	292 × 584	3,212 × 1,732	1,284 × 1,343	1,868	1,343 × 1,460	9,928	11 mm.	4,672	76(?)	116-525	58-120	
	31 × 14 × 2.3	467	700 × 700	? × 408	2,336 × 2,044	876 × 1,168	1,868	642 × 876	7,884	13 mm.	4,555	45	116-700	58-175	
	35 × 9	378	344 × 498	* 292 × 516	2,732 × 2,150	..	..	1,204 × 1,720	7,740	..	5,160	67	175-584	58-116	
	43 × 12	326	550 × 584	* 326 × 430	2,924 × 2,201	..	..	1,548 × 1,548	9,718	..	5,160	53	..	..	Slightly irregular rounded
	16 × ?	..	395 × 481	* 86 × 172	1,874 × 1,100	..	..	860 × 820	4,644	..	2,408	52	..	..	Sagittal section
F. rath.	40 × 9 × 2	292	498 × 567	51 × 172	2,854 × 1,823	..	..	946 × 1,376	8,944	..	4,816	54	..	..	
	15 × 10	..	..	* 408 × 467	2,336 × 1,752	584 × 1,460	..	700 × 876	..	7 mm.	..	..	175-408	..	Cirrus protruding
	21 × 11	292	467 × 700	116 × 350	..	292 × 1,284	..	759 × 1,168	4,263	9 mm.	..	..	..	58-175	
	19 × 11	292	642 × 817	175 × 350	? × 2,044	876 × 1,401	..	516 × 688	4,568	..	2,401	56	..	58-116	
F. goddardi	16 × 7	120	309 × 447	34 × 120	1,892 × 1,513	..	..	946 × 1,720	4,588	..	4,128	83	..	..	Sagittal section, greatly contracted
	20 × 12	..	464 × 430	..	2,476 × 1,462	..	..	..	..	..	..	..	..	..	
	22 × 10	292	..	175 × 408	2,452 × 2,044	525 × 1,168	1,168	1,168 × 1,284	4,672	9.5 mm.	2,628	56	? - 584	58-175	
	23 × 10	292	..	235 × 407	2,531 × 2,044	642 × 1,401	1,051	1,460 × 1,460	5,256	11 mm.	3,679	69	? - 408	..	
	22 × 11	292	..	..	2,044 × 1,635	817 × 1,109	..	876 × 876	6,716	10 mm.	..	..	116-350	58-116	
	20 × 6	..	..	..	2,336 × 1,752	580 × 700	1,284	934 × 1,343	6,192	10 mm.	3,796	55	116-350	..	
	20 × 6	..	..	..	2,201 × 1,444	..	..	946 × 1,290	6,192	..	2,408	40	..	..	Cirrus protruding
	23 × 9	240	619 × 636	* 172 × 335	2,580 × 1,806	..	..	946 × 1,118	5,160	..	3,658	51	..	..	Sagittal section, cirrus protruding
	23 × ?	86	395 × 567	* 108 × ?	2,322 × 1,462	..	..	..	..	..	3,440	66	..	..	Slightly heart shaped
	23 × 8	344	..	* 154 × 516	2,081 × 1,840	..	..	1,118 × 1,118	4,472	..	3,010	68	..	..	Sagittal section, contracted
F. goddardi	23 × 8	103	584 × 705	64 × 206	2,938 × 1,720	..	..	946 × 1,032	4,128	..	1,892	47	..	..	
	23 × 10	..	464 × 447	..	2,150 × 1,462	..	..	1,118 × 1,238	7,740	..	3,612	46	..	..	
	20 × 6	223	516 × 567	* 154 × 240	2,304 × 1,892	..	..	946 × 1,204	4,816	..	2,408	50	..	..	

\* First column is length of sac posterior to acetabulum; second column is ratio of this length to entire distance between acetabulum and shell gland.



Measurements of the various organs are tabulated in Table 1, from which it appears that the variations which occur are not characteristic of the groups, but rather bear a general relation to the size and development of the individual.

6. *The Ova*.—Observation of the ova in hundreds of samples of fresh feces leaves an impression on one of their essential unity, an

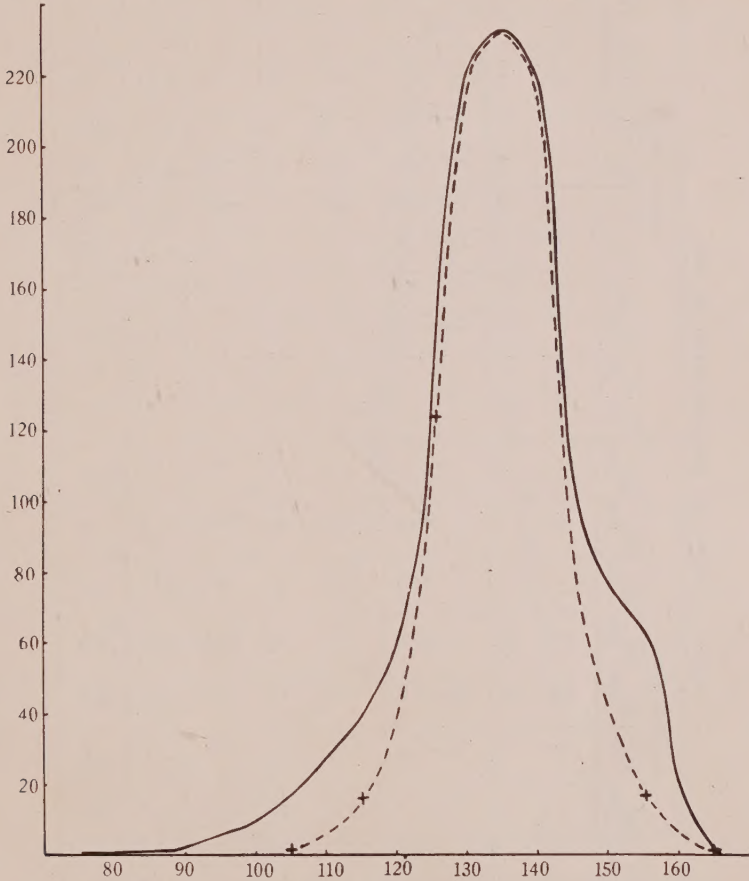


Fig. 4. Curve representing length of ova taken from entire series.

impression confirmed by the present series of measurements. In all, 576 eggs were measured, of which 150 were from two separate samples of fresh feces, and the rest from the lower uteri of nine preserved flukes, including representatives of each variety. The entire series is found to be grouped, in length, about the single mode 130-140 $\mu$  with variations conforming fairly well to the normal curve (Graph 4). The same mode, it will be noted, obtains for each of the groups as well,

TABLE 2.—TABLE OF MEASUREMENTS OF OVA

Source	No.	Length in Microns										Width in Microns										Remarks			
		71/ 80	81/ 90	91/ 100	101/ 110	111/ 120	121/ 130	131/ 140	141/ 150	151/ 160	161/ 170	51/ 55	56/ 60	61/ 65	66/ 70	71/ 75	76/ 80	81/ 85	86/ 90	91/ 95	96/ 100		101/ 105	106/ 110	
F. buski	1. 40×11.....	...	...	...	...	21	58	...	...	...	...	...	...	...	...	...	...	4	9	42	12	6	6	Distortion, swelling, asymmetry present	
	2. 27×9.....	...	...	...	...	2	16	10	...	...	...	...	...	...	...	...	16	0	0	2	0	0	1		
	Group b.....	...	...	...	...	2	37	68	...	...	...	...	...	...	...	...	4	9	44	12	6	7			
	F. rathousi	30	...	...	4	8	18	...	...	...	...	...	...	1	5	3	10	5	5	1	...	...	Some swelling Slight asymmetry		
	30	...	...	...	...	14	16	...	...	...	...	...	...	...	...	...	20	9	1	...	...	...			
Group r.....	5. 17×9.....	...	...	...	...	3	15	...	...	...	...	...	...	...	...	...	6	0	2	1	...	...	General swelling, frequent asymmetry		
	6. 22×13.....	...	...	...	...	...	51	38	10	...	...	...	...	...	...	...	50	41	9	2	...	...			
	100	...	...	...	...	25	47	66	10	...	...	...	...	...	...	...	96	55	17	2	...	...			
	F. goddardi	30	1	1	5	14	7	2	...	...	...	...	...	1	...	...	2	3	4	9	4	2		General swelling, frequent asymmetry	
	50	...	...	...	...	...	1	...	...	...	...	...	...	...	...	...	24	19	3	4	...	...			
Group g.....	49	...	...	...	...	...	4	12	32	1	...	...	...	...	...	...	8	14	20	7	...	...	Nos. 7 & 9 omitted		
	...	1	1	5	14	7	3	11	36	50	1	1	...	1	...	...	34	36	27	20	4	4		2	
	Feces	50	...	1	...	1	5	28	13	2	...	...	...	...	4	...	11	31	...	2	...	2		Nos. 7 & 9 omitted	
	2. Sept. 1917.....	100	...	1	1	6	19	92	27	4	...	...	...	...	...	...	76	54	8	2	...	2			Nos. 7 & 9 omitted
	Group f.....	...	...	1	1	...	...	...	...	...	...	...	...	...	...	...	...	...	...	...	...	...			
Total.....		576	1	6	19	40	106	237	101	64	1	1	...	3	16	19	222	149	61	68	16	12	9	Nos. 7 & 9 omitted	
Series 2.....		497	...	1	5	33	104	233	89	32	...	...	...	2	16	19	212	132	37	50	12	8	7		
Group 2		...	1	5	21	17	57	68	...	...	...	1	0	2	7	10	28	12	18	54	16	10	9	Nos. 4, 5, 6, 8, 9.....	
Nos. 1, 2, 3, 7.....		...	...	...	...	17	30	77	...	...	...	...	...	...	...	...	6	118	83	35	12	...	...		



except Group G, in which it is raised somewhat by the abnormality discussed below. If now on account of the great divergence shown by individuals in this group and for other reasons we throw out *F. goddardi* altogether, and make but two species as shown in Group 2 of the table (Table 2), here again both species—*buski* and *rathouisi*—

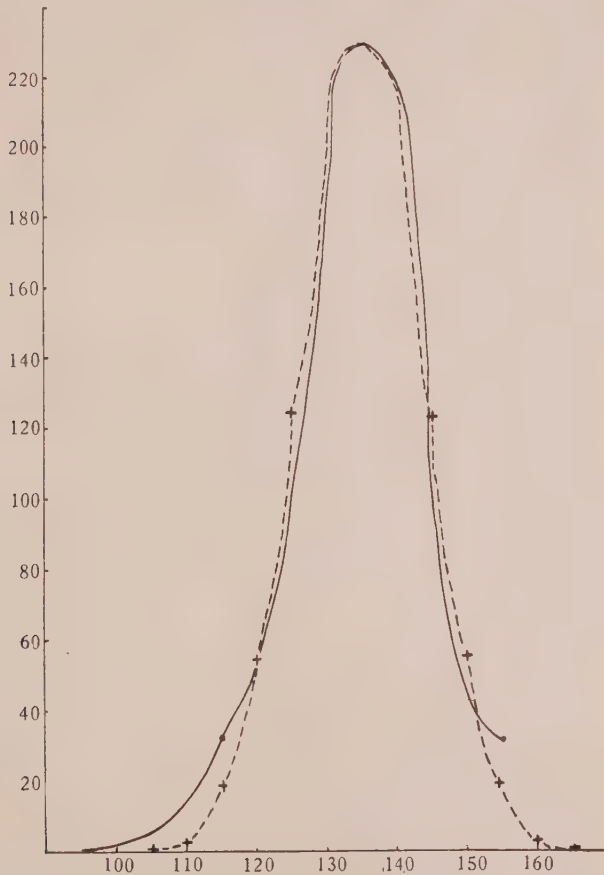


Fig. 5. Curve representing length of ova in amended series.

are found to have the same mode. It is also noteworthy that eggs deposited in feces—a normal condition—form a curve which agrees very closely both with its own normal and with the curve for the entire series, and has a much smaller standard deviation—spreads less—than do any of the curves for eggs from an individual. Graphs 4-7 graphically represent this fact, which is strikingly apparent under the microscope, viz., that ova taken from the body of the parasite present

far greater variations in contour and in size than are seen under normal conditions. In particular, such variations in contour were especially noticeable in eggs from fluke 9, and a large proportion of them were apparently swollen; i. e., a clear space appeared between the yolk contents and the shell, the former remaining normal in size and shape, while the shell measured from  $23\text{-}27\mu$  more in length, and about 11 more in width. To a less extent these conditions were found also

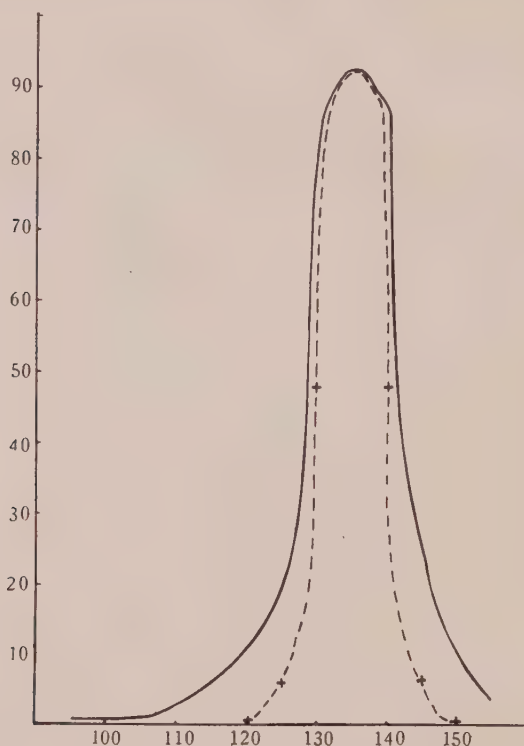


Fig. 6. Curve representing length of ova taken from feces.

in flukes 2, 3 and 5, and are doubtless abnormalities, as I have never seen them in feces specimens. On the other hand, eggs from fluke 7 were relatively so few (i. e., the total number obtained from the uterus were so few), and are grouped about a mode so much smaller than the average that they may fairly be considered immature, especially since in another fluke of about the same dimensions practically no eggs were found. If now on the supposition that they are abnormal, the eggs from these two flukes, 7 and 9, be excluded from the series, and the curve of the remainder be plotted as is done in Graph 5, the close correspondence between observed and theoretical curves furnishes



conclusive evidence of the correctness of the hypothesis. Further, that these variations are individual abnormalities rather than indications of specific differences seems clear from the fact that in the entire series they stand alone, that the flukes from which they were taken are in other respects indistinguishable from each other and from flukes yielding normal eggs, and that the eggs differ as described above from any that I have seen that were deposited in the normal manner. In the above discussion the short diameter of the egg has not been considered because it does not alter the problem. Those measurements, however, will all be found in the table (Table 2).

Writing in the *Encyclopaedia Britannica*, Peter Chalmers Mitchell defines a species as "an assemblage of organic forms which . . . if they differ among themselves differ less markedly than they do from

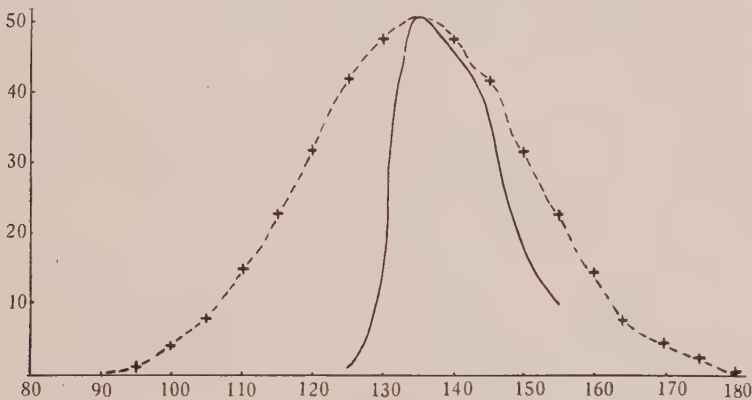


Fig. 7. Curve representing length of ova taken from fluke No. 6.  
For further details compare text.

those outside the species, or if differing markedly are linked by intermediate forms." With such a definition and in the light of the foregoing discussion it would seem that the three varieties represented in the Shaohing specimens should be regarded as belonging to but one species, *F. buski*. Poirier's original specimen on which the species *rathouisi* was based, came from a suburb of Shanghai, the commercial metropolis of the region, to which people from neighboring cities have flocked in large numbers. In recent years a few cases of fasciolopsis infection have been reported from Shanghai, and some of them at least are known to have been natives of Shaohing. It is therefore quite probable that Poirier's specimen came from Shaohing, which if true would tend to support the argument from morphology presented by Odhner and others to the effect that it was in reality but a contracted form of *F. buski*. Kwan's fluke has been conclusively shown by Leiper to be a mutilated specimen of the same species, and the

description of *F. fülleborni*, given by Rodenwaldt, agrees with that of *F. buski*, as modified by the present study in every particular except that it is said not to possess cuticular spines nor the characteristic diverticulum of the seminal vesicle. If Leiper's statement, already quoted, regarding spines in this case be accepted, there remains but a single point of difference outstanding; and in view of the limited amount of material (three specimens from a single case) on which the description was based, further study would seem to be required before the identity of this as a separate species can be accepted.

#### MORPHOLOGY OF THE PARASITE

Assuming then the existence of but a single species in the Shaohing flukes, the following account of its morphology may be given, which applies equally to all varieties, except that measurements of internal parts are to be understood as applying to individuals approximating the average in size.

*General Appearance.*—*F. buski* is a flat, elongated fluke, typical specimens measuring about 30x12x2 mm. Busk reported a maximum length of three inches (75 mm.?). In Shaohing the greatest measured length was 55 mm., and the greatest width 20. Immature forms 5 mm. or less in length have been recovered from feces, and in six uncontracted forms measuring from 9 to 21 mm., but a few poorly developed eggs were found in the longest, and in the others none at all, from which the minimum adult length may be assumed to be 20 mm.

The head end is somewhat pointed, with a poorly defined shoulder, the tail bluntly rounded, the lateral margins even or slightly wavy. In fresh specimens the color is a uniform deep pink—ham color—and to the touch and the unaided eye the integument appears smooth with transverse lines more or less prominent according to the state of muscular contraction.

*The Cuticula.*—Microscopically, this is seen to consist of a basement membrane overlaid with the true cuticula, which under high magnification shows fine striae vertical to the plane of the surface. It is somewhat thicker on the dorsal (19 to 34 $\mu$ ) than on the ventral surface (15-27), the maximum being at the level of the upper border of the acetabulum, from which point it becomes thinner in both directions. True cuticula similar to that covering the surface of the body, but thinner, extends for varying distances into all the organs which open externally.

The entire ventral surface, including the narrow strip anterior to the oral sucker, is armed with spines, which are most numerous in the acetabular region (Pl. XVII, A). These spines are deeply imbedded in the cuticula, their bases resting on or in the basement membrane,



and are directed caudad at an angle of 30-45 degrees, the merest tip projecting beyond the surface. They are arranged, somewhat irregularly, in alternating transverse rows (Pl. XIII, *B*) which in the most thickly covered regions may be almost in contact, but will average 10 to  $15\mu$  apart, and toward the tail may be separated by  $160\mu$  or more. Similarly, in the transverse direction the spines are practically in contact over the acetabulum, but toward the tail the interval between the bases of adjoining spines varies from 19 to  $40\mu$ .

*The Spines.*—The spine itself (Pl. XIII) is a scale-like structure, 25 to  $30\mu$  wide at the base, and 30 to  $34\mu$  long. The sides converge a trifle toward the tip which is bluntly rounded and curved backward. The upper surface of the spine is flat or, especially toward the tip, slightly concave. In the lower surface the curvature is somewhat greater, giving to a transverse section a crescentic outline. The body of the scale increases in thickness from a thin edge at the tip to 10 to  $13\mu$  at the base where it flares out rapidly like the thorn of a rose. The surface of the base, where it is attached to the basement membrane, presents two, three or rarely four cusps, causing a transverse section through it to appear like a group of rounded or irregular bodies. The size of the scales varies with the stage of development of the parasite, and with the location, being smallest around the oral sucker and the genital pore. Extremes in length were 22.8 and  $41.8\mu$ , other dimensions being in proportion.

*The Musculature.*—Muscular fibers occur in the walls of the various viscera, and are particularly well developed in the cirrus and the metratrum. In addition, the muscular system comprises the following:

1. *The dermo-muscular tube* lies immediately beneath the basement membrane of the cuticula, and consists of annular, oblique and longitudinal fibers. At rather frequent intervals are found other longitudinal and also dorso-ventral fibers (the parenchymal muscles), and certain fibers attaching various viscera, especially the two suckers and the shell gland to the dermo-muscular tube.

2. *The oral sucker* is situated at the anterior extremity, its orifice being sub-terminal on the ventral surface, and its long axis, which is continuous with that of the pre-pharynx and pharynx, being oblique to the surface (Pl. XII, *B*). In younger specimens the sucker is nearly globular, but with age the transverse diameter becomes longer, and the dorso-ventral shorter relative to the longitudinal. In well developed specimens these diameters will range from about 0.5 to 0.7 mm. The oral orifice is normally circular, 0.3-0.4 mm. in diameter. The entire organ (and this applies as well to the pharynx and the ventral sucker) is enclosed in a capsule and suspended by means of several processes

of muscular and connective tissue in a sinus of the excretory system, doubtless in order to facilitate muscular contractions. In addition to this, considerable motion in its longitudinal axis, whereby the organ may be partially extruded through the oral ring, is made possible by its free attachment to the ring by means of an eversible collar of cuticula and connective tissue, and by the interposition of a collapsible portion of the alimentary tract—the pre-pharynx—between the oral sucker and the pharynx. Whether this freedom of motion is used for locomotion as well as for feeding is not determined, but seems probable.

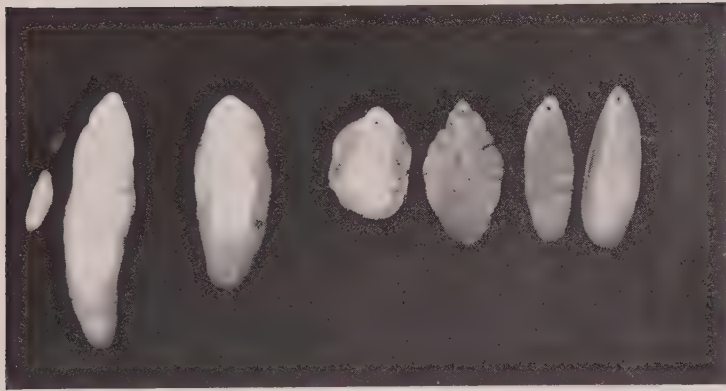
2. *The pre-pharynx*, whose function seems to be to permit motion of the oral sucker, is surrounded by a well developed muscle consisting of circular fibers, and extending from the upper surface of the pharynx to about the middle of the oral sucker, which it is thus able powerfully to reinforce (Pl. XII, *B*).

3. *The pharynx*, which surrounds the alimentary tract just prior to its bifurcation and lies beneath the dermo-muscular tube posteriorly, is a spheroidal mass of radial and circular fibers measuring from 0.40 to 0.75 mm. antero-posteriorly, and from 0.70 to 0.99 mm. transversely.

4. *The ventral sucker* is a powerfully developed, bell-shaped organ, situated near the anterior extremity (1.2 to 1.8 mm. from the tip) and so placed that its long diameter and the plane of its orifice are oblique to the ventral surface, the anterior lip of the orifice being somewhat longer than the posterior. Its total length varies from 2.3 to 3.2 mm., and its greatest diameter from 1.7 to 2.5 mm., the dorso-ventral diameter being somewhat shorter than the transverse. The orifice, normally circular with a diameter of 1.0 to 1.5 mm., is in preserved specimens often transversely elliptical or irregularly heart-shaped. The acetabular cavity is lined throughout with an extension of the cuticula.

*The alimentary tract* extends as a single tube from the oral aperture to just beyond the pharynx where it bifurcates, forming the two ceca which proceed laterally to the outer margins of the acetabulum, at which points they bend sharply caudad and follow a sinuous course to the posterior extremity, ending blindly near each other. This course is marked by two main inward curves, viz., at the level of the shell gland, and between the testes; but the ceca follow the outline of the viscera they enclose and hence the number and degree of their curves depend in part upon the development of the individual, and the state of its muscular contraction. The lumen of this tract varies considerably in size. At the oral aperture it appears in sections as a transverse slit, 0.2 to 0.3 mm. by 0.085 to 0.100 mm., but immediately expands, then narrows, to expand again within the pharynx, from





*A*      *e*      *f*      *g*      *i*      *j*      *k*



*B*

## PLATE XII

### EXPLANATION OF PLATE

A. Varieties of the fluke encountered in Shaohing.

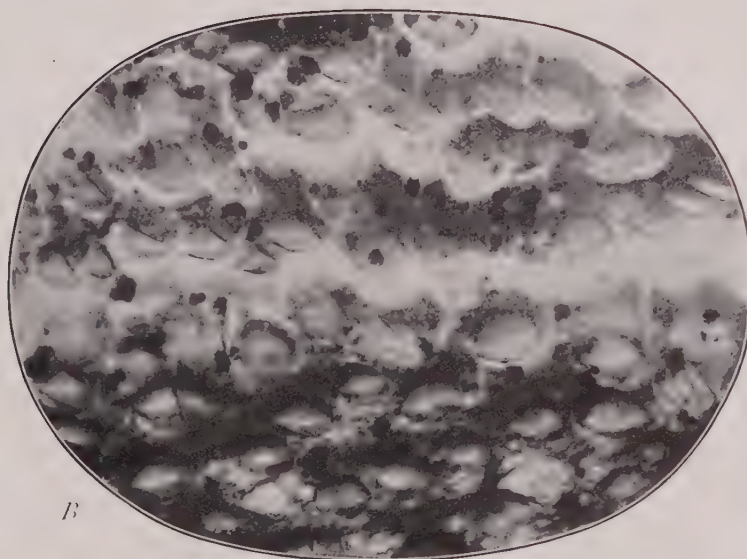
B. Anterior extremity, showing especially the sinuses surrounding oral sucker, pharynx, and ventral sucker; nerve cell in pharynx; cuticula stripping off and also lining of oral and ventral suckers. Sagittal section. ( $\times 68$ .)







A



B

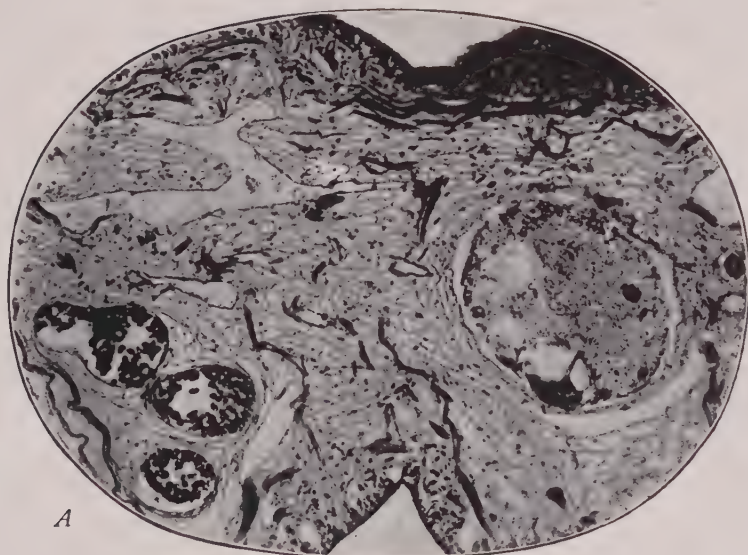
PLATE XIII

EXPLANATION OF PLATE

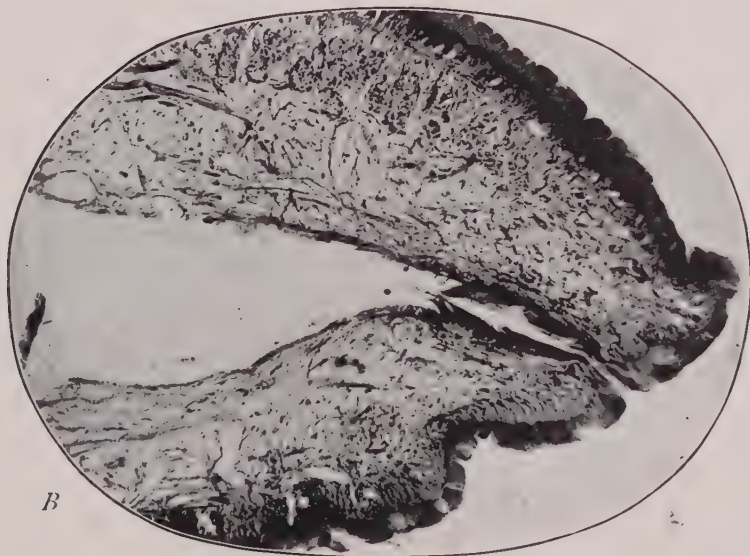
A. Spines embedded in cuticula, and being stripped off with it. Sagittal section. ( $\times 450$ .)

B. Ventral spines, arranged in alternate rows. Section in ventral plane. ( $\times 450$ .)





A



B

PLATE XIV

EXPLANATION OF PLATE

A. From fluke F showing entire absence of cuticula. Also coils of ovary at left, nerve cell at right. Sagittal section. ( $\times 68$ .)

B. Posterior extremity, showing excretory vesicle and duct. Sagittal section. ( $\times 68$ .)





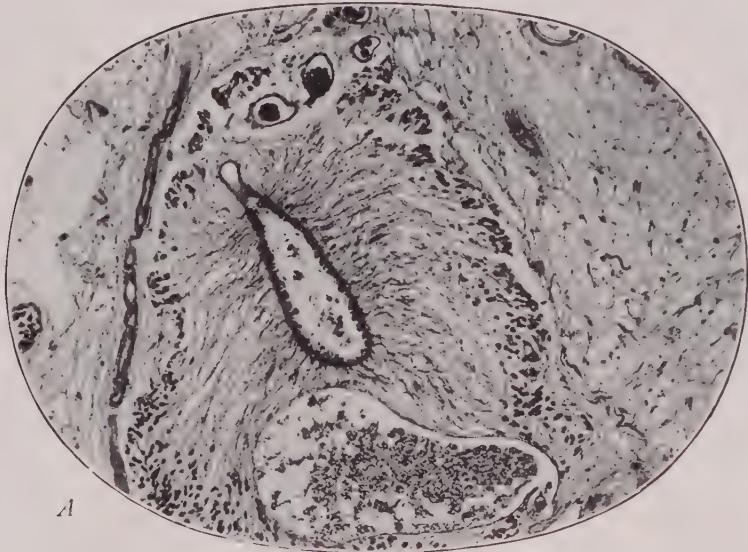


PLATE XV

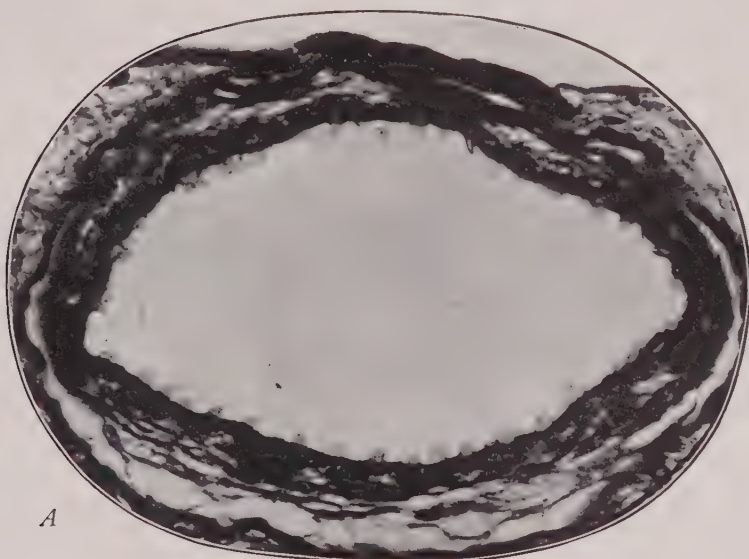
EXPLANATION OF PLATE

A. Shell gland, with vas efferens at left, coils of Laurer's canal above, junction of common yolk duct with oötype in center, and vitelline receptacle below. Ventral section. ( $\times 90$ .)

B. Metratrum and everted cirrus just below level of genital pore, showing 3 large spines in metratrum, also spines of cirrus and pre-cirral canal. Ventral section. ( $\times 360$ .)







A



B

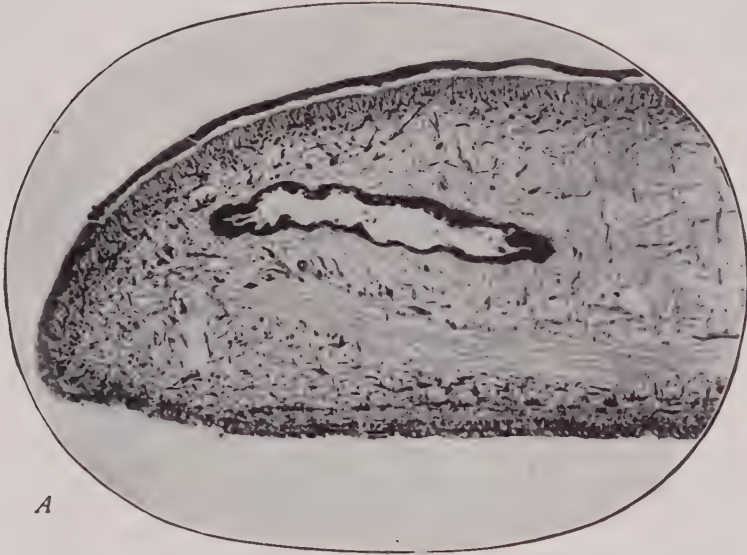
PLATE XVI

EXPLANATION OF PLATE

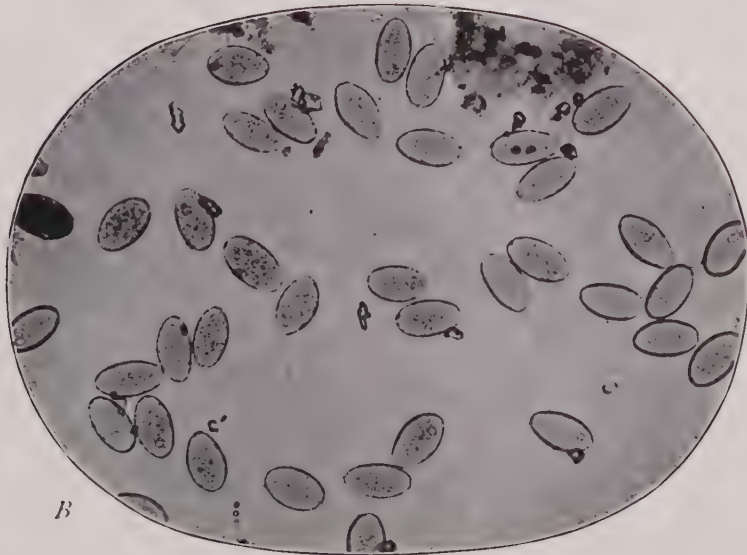
A. Pre-cirral canal, showing spines. ( $\times 450$ .)

B. Three nerve cells at base of oral suckers and others in pharynx. Note how freely pharynx is suspended in sinus of excretory system. ( $\times 90$ .)





A



B

PLATE XVII

EXPLANATION OF PLATE

A. Sagittal section, showing nerve trunk at level of ventral sucker with a nerve cell near anterior extremity. Cecum at right. Note cuticula with spines on ventral surface, stripping off on dorsum. ( $\times 45$ .)

B. Group of eggs from feces. ( $\times 68$ .)





which it emerges as a tube 0.135 to 0.175 mm. in diameter. This portion of the tract, the esophagus proper, is very short (85 to 100 $\mu$ ), as the transverse portion of the ceca lies practically in juxtaposition with the pharynx. The diameter of the ceca is not constant, but at the acetabulum approximates 230 $\mu$  and diminishes somewhat toward the tail. Up to the point of its bifurcation this canal is lined with an extension of the cuticula; beyond that point with tall columnar epithelium arranged in several (5-9) longitudinal ridges, which give to it a characteristic stellate appearance on cross section.

*The excretory system* begins immediately under the basement membrane of the cuticula as small spaces or canals which coalesce to form larger ones, and eventually empty into the excretory vesicle. In the anterior part of the body, three main trunks—a central and two lateral—gather up the smaller branches and unite at the lower border of the shell gland to form the excretory vesicle, which occupies the central portion of the fluke from this point to the extremity, with a diameter about one third of the thickness of the body at that part—approximately 500 or 600 $\mu$ . Throughout its extent it receives transverse and oblique tributaries, and from its caudal extremity a short, straight duct, 10 or 12 $\mu$  in diameter, leads to the external orifice on the posterior surface, about 300 $\mu$  from the tip (Pl. XIV, B). This excretory pore is provided with a sphincter in the dermo-muscular tube; the duct is lined throughout with cuticula, the vesicle with tall columnar epithelium, and the other branches of the system with a single layer of flattened cells.

Striking features of the anatomy of the fluke are the large sinuses surrounding the oral sucker, the ventral sucker, the pharynx, and to a less extent the shell gland and other organs, and communicating directly and freely with the main trunks of the excretory system. It seems reasonable to suppose that in addition to their excretory function these sinuses are analogous to the synovial or pleural sacs of mammals (Pl. XII, B, and Pl. XVI, B).

*The Male Reproductive System.*—The testes, two in number, consist of thick tubes, 0.6 or 0.7 mm. in diameter, and about one third the width of the fluke in length, which lie one behind the other ventral to the excretory vesicle, and with their branches occupy the greater part of the posterior half of the body. The branching is frequent and usually dichotomous, the finest divisions ranging from 116 to 200 $\mu$  or more in diameter. From each main trunk a vas deferens arises, and these pass forward over the surface of the shell gland on either side (Pl. XV, B), and converge to pierce the cirrus sac and empty into the posterior tip of the primary seminal vesicle. This point, the beginning of the cirrus sac, is usually easily made out in cleared specimens, and

lies ordinarily about midway between the shell gland and the tip of the acetabulum, but may be very much nearer the former.

The cirrus sac consists of muscular and fibrous tissue, and contains from behind forward the following structures: seminal vesicles, ejaculatory duct, cirrus and pre-cirral canal, the last three of these constituting the vas deferens. The first portion of the sac terminating at the tip of the acetabulum is more or less convoluted, and has a diameter of 500-700 $\mu$ . The second portion containing the vas deferens follows a straight course longitudinally, lying in apposition with the acetabulum in front, and with the metraterm to the left. Anteriorly, its walls blend with those of the genital atrium.

The seminal vesicles are two more or less convoluted tubes, lying side by side within the first portion of the cirrus sac. One of these vesicles, here termed the primary vesicle, extends caudad slightly farther than the other and receives the vasa efferentia. Its distal extremity empties into the lumen of the secondary vesicle (the so-called cecal appendage of the former descriptions) where it narrows to form the ejaculatory duct, but is directed caudad. Thus both vesicles are practically always full of sperm. Both vesicles are lined with a cylindrical epithelium.

The spermatozoön possesses the usual form—namely, a spindle-shaped body and a filamentous tail, with a total length of 6 or 7 $\mu$ . The ejaculatory duct, approximately the first half of the vas, is continuous with the seminal vesicle behind and the cirrus in front, and differs from the former only in being straight and smaller in diameter (140-200 $\mu$ ): The cirrus forms about half of the remaining portion of the vas. The muscular fibers in its walls are notably developed and it is lined with very small and delicate spines, which hardly take the stain for chitin (Pl. XV, B). The distal extremity of the cirrus protrudes into the pre-cirral canal very much as the human cervix uteri extends into the vagina.

The pre-cirral canal which opens anteriorly into the base of the genital atrium, is closely set throughout with spines similar to the cuticular variety, but much smaller and more delicate, being 6.5x7 $\mu$  (Pl. XVI, A). When the cirrus is protruded this canal is entirely evaginated, forming then an outer coat enclosing the whole length of that organ except its tip. This doubtless will account for the statement by Stephens that the cirrus "is beset with very fine spines except at either extremity"; for the spines in the pre-cirral canal do not extend over the portion of the cirrus which projects into it (Pl. XV, B).

The genital atrium, into the bottom of which the pre-cirral canal and the metraterm empty side by side, is 250 to 300 $\mu$  deep, and when relaxed appears as a transverse slit some 400 to 500 $\mu$  wide. During



extrusion of the cirrus, however, that organ completely fills the lumen, and considerably distends it, giving it a cylindrical form. The atrium is lined with cuticula, but without spines.

*The Female Reproductive System.*—The vitelline glands are symmetrical structures, occupying the lateral portions of the fluke, and extend from the level of the acetabulum to the posterior extremity where they meet in the middle line. They lie superficially, immediately within the dermo-muscular tube, extending inward on both the ventral and dorsal surfaces for about one sixth of the width of the fluke, and thus enwrap to a certain extent the outlying parts of the testes, uterus, etc. Their acini are round or oval, and lined with large celled epithelium (18 to  $20\mu$ ), with prominent round nuclei (4 to  $5\mu$ ) and numerous refractile granules (2 to  $2.5\mu$ ). On each side the yolk substance is collected by numerous tubules into an anterior and posterior longitudinal tubule, which unite to form the transverse yolk duct,  $100\mu$  in diameter. When near the shell gland the transverse ducts bend dorsalward to enter it in its dorsal and posterior portion, and expand at their junction within it to form the vitelline receptacle (Pl. XV, *A*), from which the common yolk duct, a small tubule, extends upwards and unites with the short oviduct to form the oötype.

The shell gland is an elastic body surrounded by a capsule of connective tissue, into which the parenchymal muscles are more or less blended. It is generally ovoid in shape with the long axis oblique to the midline of the body, the anterior end to which the ovary is attached being to the right of this line. In the smaller flukes it is situated but a millimeter or less anterior to the mid point of the body, but in the longest specimens lies at about the junction of the anterior and middle thirds. The cells of this gland are generally spindle-shaped ( $10 \times 22\mu$ ), but some are rounded (10 to  $15\mu$  in diameter) with large, round, deeply staining nuclei ( $5\mu$  in diameter). These are gathered in a peripheral zone  $100\mu$  or more in thickness, from which delicate processes converge toward the oötype.

The ovary consists of three stems—superior, middle and inferior—the outer ends of which are closely branched, while inwardly they soon merge into a common mass lying upon the upper right hand portion of the shell gland near the ventral surface. From this mass a small duct, the oviduct, sinks into the shell gland, takes a convoluted course to the posterior part of the gland where it turns toward the opposite side, gives off Laurer's canal, and immediately afterwards is joined by the common yolk duct to form the oötype. The cells of the ovary, or ova proper, stain a dark reddish brown with van Gieson, are generally nearly round (12 to  $15\mu$  in diameter), with large, round, deeply staining nucleus (7 to  $8\mu$ ) and nucleolus ( $5\mu$ ).

The oötype is a straight cylindrical tube, 100 to 135 $\mu$  in diameter, extending transversely through the substance of the shell gland, and is continuous at either end with the oviduct and the uterus, respectively. It is lined with a layer of tall columnar cells lying upon a basement membrane, and in stained sections is surrounded to a depth of 50 to 60 $\mu$  with deeply stained, radially arranged lines—the terminal portions of the processes of the cells of the shell gland—which give to it a characteristic caterpillar-like appearance.

Laurer's canal is a small duct given off from the oviduct just before that canal unites with the common yolk duct to form the oötype. It follows a convoluted course, especially in that part within and adjacent to the shell gland, and empties on the posterior surface in the mid line at a point a millimeter or so in a straight line from its origin. It is lined throughout with an extension of the cuticula, and is provided with a sphincter at its distal extremity. In general, its lumen is about 12 $\mu$ , but frequent enlargements or sacculations of the tube occur, having twice or three times that diameter. There is no trace of any receptaculum seminis. Sperm was not found in any of the cases examined, but only a few cells from the ovary, and still more rarely yolk cells. Probably these were abnormally forced into the canal during the death spasm.

The uterus in the first part of its course is differentiated from the oötype by the absence of the caterpillar-like rays, and by its convoluted course. It leaves the shell gland near the upper left hand portion of its ventral surface, and arranged in loose coils occupies most of the anterior portion of the fluke as far as the acetabulum. In the larger flukes it is packed with eggs, and may attain a diameter of 500 $\mu$  or more. The first portion of the uterus is lined with cylindrical epithelium—as is also the oviduct—but in the central portion this is reduced to a layer of thin, flattened cells with delicate processes projecting into the lumen like spines in appearance except that they do not stain. At the tip of the acetabulum the uterus merges into the metraterm, becoming reduced in size (about 150 $\mu$ ), the walls are thickened, muscular fibers especially being increased and the lining becomes identical with the outer cuticula, and is armed with stout spines measuring approximately 23x28 $\mu$  (Pl. XV, B). These have been found embedded in the cuticle from the outer termination of the metraterm for somewhat over two thirds of its length and may possibly normally be found throughout its entire extent. Throughout its course the metraterm lies upon the acetabulum in juxtaposition with the vas and to its left.

The ovum is an ellipsoid, rather bluntly rounded at either pole, and measuring normally 130 to 140 $\mu$  by 80 to 85 $\mu$ . The shell is clear and

thin, the operculum so delicate as to be made out only with difficulty in many instances, and the contents consist of a large number of yolk cells with usually but a single germinal cell situated towards the operculum from the center. Considerable variations in size and shape are met with, though they are not common in eggs deposited in the normal manner, as is apparent from a study of Table 2, Graphs 4, 5, 6, and Pl. XVII, *B*; from these it also appears that the normal egg as found in feces is not so pointed toward the poles as it has been figured hitherto.

*The Nervous System.*—The nervous system may be divided into a peripheral and a visceral portion. The peripheral portion consists of a group of numerous ganglion cells clustered around the lower portion of the oral sucker (Pl. XVI, *B*), from which three nerves proceed on each side.

1. The ventral nerves pass rapidly forward toward the surface, and from the level of the acetabular orifice to their termination at the posterior extremity are found immediately below the dermo-muscular tube and slightly within the general course of the ceca (Pl. XVII, *A*). Anterior to the acetabulum they are united by three commissures, one at the base of the pharynx, and the others respectively above and below the transverse portion of the ceca. Throughout the remainder of their extent at intervals of from 300 to 700 $\mu$  they give off median and lateral branches which, with corresponding branches from the other nerves, are distributed to the entire periphery. Opposite the acetabulum the ventral nerves are 135 $\mu$  or more wide and somewhat less in thickness, and taper gradually toward the tail.

2. The dorsal nerves are superficial throughout their entire course, are united by a single commissure behind the cecum, and are somewhat smaller than the ventral nerves, but similar in their branchings and distribution.

3. The lateral nerves arise from the common origin and by a plexus of three or four branches from the ventral nerves on either side, and are distributed to the shoulders, terminating at about the level of the acetabulum.

Small bundles of fibers from the ventral nerves have been traced to the acetabulum and pharynx, and doubtless the two portions of the nervous system are intimately correlated, though this has not been demonstrated. But in general it is evident the portion of the system just described is concerned primarily with the innervation of the periphery, including the dermo-muscular tube.

4. The cells referred to as "ganglion cells" above, are elliptical cells about 38 $\mu$  long, and 30 $\mu$  in their short diameter. The nucleus is large and round; and the cytoplasm finely reticulated. In some

instances fibers have been seen to pass from the cell into the nerve bundle. As stated above, a large group of these cells is to be found around the base of the oral sucker, but single cells occur also at various points along the course of the nerves.

The visceral portion of the nervous system consists of round or oval cells similar to the ganglion cells of the peripheral portion, but without visible fibers leading from them. They occur in great numbers throughout the body, being most numerous in the powerfully developed muscles (oral sucker, pharynx, and ventral sucker), and in the cirrus and the metraterm; and to a less extent about the other viscera.

The foregoing detailed description may be summarized as follows: *Fasciolopsis buski* is a flat, elongated fluke, presenting great variation in size, but averaging about 30x12x2 mm. It is deep pink in color, and surface appears smooth except under microscope when ventral surface is seen to be beset thickly with spines. Oral sucker sub-terminal on ventral surface; ventral sucker powerful and near anterior extremity. Genital pore just anterior to ventral sucker. Vitellaria are racemose glands occupying lateral portions from ventral sucker to tail. Shell gland oval in mid line of body somewhat anterior to mid point. Uterus in loose coils occupies anterior portion of fluke, its terminal portion, the metraterm, being spined and emptying into the common genital atrium beside the pre-cirral canal. Ovary, closely branched, attached to shell gland on the right. Testes, two in number, closely branched, and lying one behind the other, occupy most of the body posterior to the shell gland. Cirrus and pre-cirral canal lie parallel with metraterm, and are armed with fine spines. Excretory vesicle large, with many transverse and oblique branches, and empties on dorsal surface, near posterior extremity.

#### CONCLUSIONS

1. Infestation with *Fasciolopsis buski* is to be regarded as a serious disease; and where local conditions favor it becomes of considerable importance from the standpoint of public health.

2. The most notable symptoms are general weakness, diarrhea, anemia, and edema. The rapid accumulation of fluid in the body may be accompanied by a pronounced suppression of urine without evidence of renal involvement.

3. Contrary to certain authorities, fever is not noted, except in complicated cases.

4. The parasite shows great variation in morphology, but withal such gradation in variation as to justify including the forms now described as *F. rathousi* and *F. goddardi* in the species *F. buski*. On



account of the close similarity of *F. fülleborni*, it would appear desirable to subject this species also to further investigation.

5. Ventral cuticular spines are characteristic of *F. buski*, and probably of the entire genus.

6. Cirrus and metraterm are spined in *F. buski*, and therefore Railliet's description of the family Fasciolidae, if it is still to include Fasciolopsis, needs to be revised accordingly.

#### ACKNOWLEDGMENTS

Grateful acknowledgement should be made here of my indebtedness to the China Medical Board of the Rockefeller Foundation for the grant of a fellowship without which this investigation would have been impossible, and to Prof. E. E. Tyzzer of the Department of Comparative Pathology of Harvard University under whose direction it has been carried on, and whose cordial interest in the work has been manifested in many helpful ways. It is a pleasure to express my indebtedness also to my colleague, Dr. C. H. Barlow, of Shaohing, China, for aid in securing some of the material; to Prof. H. B. Ward of the University of Illinois for helpful suggestions, and to Prof. F. B. Mallory of Harvard University, and his able assistant, Miss Lillian M. Leavitt for the fine series of microphotographs which accompany this article.

## NOTES ON SOUTH AFRICAN CERCARIAE \*

ERNEST CARROLL FAUST

During the last few years Capt. F. G. Cawston of the South African Medical Corps has published several articles on South African cercariae. But, as Cort (1919:488) has so aptly remarked, "his descriptions and figures of this [cercaria of *Schistosoma haematobium*] and other forked-tailed cercariae which he has described are so entirely inadequate that it seems to me that his entire work needs verification by more competent observers." The writer has made a careful analysis of slides and alcoholics of species which Cawston has sent to Professor Henry B. Ward and presents the data in this paper.

### *Cercaria gladii* Cawston 1918 (Fig. 1)

This furcocercous cercaria, found in *Isidora schakoi* at Potchefstroom, Transvaal, is of striking interest because of its conspicuous tail, the forks of which are prolonged into long, sword-like processes. The larva measures 0.25 mm. in length by 0.073 mm. in width. The main portion of the tail is 0.29 mm. long, while the furci have a maximum length of 0.38 mm. The body is distinctly glandular and the tail is conspicuously muscular. The integument is entirely covered with minute spines. The oral sucker has a transverse diameter of  $33\mu$ . The acetabulum lies about two-thirds the body distance from the anterior end. It is only  $26\mu$  in diameter. The oral sucker opens into a short undivided gut without evidence of any pharynx. Cawston's Figure 3 (1918:69) is a very inadequate and inaccurate diagram of this cercaria. Heavy mucin ducts empty thru the ventral margin of the oral sucker, rather than farther anteriad, as in many furcocercariae. The openings are tipped with hollow piercing spines. Each of the two groups of ducts can be traced back to three large mucin glands, with relatively small nuclei and a network of granules in the cytoplasm. A mass of many germ cells is found near the posterior margin of the body. The central nervous system is unique in its position at the inner end of the gut. From it extend caudad two main ventral nerve trunks and delicate dorsals. No cyst granules have been found in the cercaria. It is quite unlikely that the larva encysts.

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\* Contributions from the Zoological Laboratory of the University of Illinois, No. 133.

Cawston has made no mention of the sporocyst of this species. Material which the writer has examined shows it to be muscular at the anterior end.

*Cercaria secobii* Cawston 1915 spec. inq.

Because of the poor mounts which have been available for study little can be added to Cawston's confusing description of *Cercaria secobii*. The data taken as a whole show clearly that this cercaria, first secured from *Physopsis africana* from the Umsindusi River, Pietermaritzburg, Natal, after the host had been subjected to an invasion of miracidia of *Schistosoma haematobium*, are not the cercariae of human bilharziosis. Cawston (1915:257) states that "the evidence is not absolutely conclusive that these cercariae were present as a result of their exposure to infection of miracidia; but, in view of the recent work done by Leiper in Egypt, the inference is allowable." Such an inference is entirely without justification either from the experimental or the anatomical data.

Our actual knowledge of this species may be summed up in the statement that it is a furcocercous cercaria with a body about 150 by 24 $\mu$ , an undivided tail trunk 200 $\mu$  long and furci equally long, developing within a sporocyst, without pigment eye-spots and probably without pharynx. It is further distinguished from the cercaria of *Schistosoma haematobium* by a somewhat smaller, narrower body and by longer tail furci. The mucin gland ducts are probably the "divided gut" of Cawston's description. The really diagnostic features of number and character of mucin glands and ducts are altogether wanting.

*Cercaria secobii* is apparently confined to the coastal region where *Physopsis africana* abounds.

*Cercaria parvocolata* Cawston 1919 (Fig. 2)

This species, found in *Physopsis africana* at Durban, Natal, has a body measurement of 120 x 50 $\mu$ , a tail trunk 220 to 250 $\mu$  long, and furci about 100 $\mu$  long. Its oral sucker is pyriform. It possesses a small, weak acetabulum, and three pairs of acidophilic mucin glands with small nuclei. A clump of germ cells is found just posterior to the mucin glands. The pair of minute eye spots is found midway between the mucin glands and the base of the oral sucker.

A most remarkable fact in connection with *C. parvocolata* is the development of the cercariae in simple sacculate rediae, which are distinguished from the usual parthenitae of the group (sporocysts) by the presence of true pharynges and rhabdocoel ceca.

*Cercaria of Schistosoma haematobium* (Figs. 3, 4, 5)

These cercariae have been described and figured by Leiper for Egypt and by Cawston and others for Natal. For the former territory *Bullinus* spp. have been found to serve as hosts for the invading miracidia, while in Natal *Physopsis africana* is the mollusk involved. The life history of this species has been thoroly established by the brilliant work of Leiper. Unfortunately, this writer has almost entirely overlooked the anatomical features of the larvae. Only the grosser features have been included in his diagrams (Leiper, 1915). These are so general as to serve no purpose in the identification of the related larval schistosome species. While the shape and relative proportions of the body, tail trunk and furci are important, items of greater diagnostic value for furcocercous cercariae are the exact type of the digestive system, including number and type of mucin glands and ducts, the number of germ-gland cells, the nervous system, and the number and relation of flame cells and excretory tubules.

Specimens of this cercaria from Natal which the writer has studied have permitted an analysis of the digestive system, including the mucin glands and ducts, and the germ cells, together with the external body features. The body of the cercaria averages about 0.24 mm. in length by 0.1 mm. in width, while the tail trunk measures 0.2 mm. long by  $47\mu$  in diameter at the base. The small blunt furci are about half as long as the tail trunk. The large oral sucker gives the larva a decidedly robust appearance. The acetabulum is small and weak. Body and tail are covered with minute spines which are heavier and longer at the anterior end of the body. The oral sucker leads into a digestive tract without any evidence of pharynx. An esophagus runs backward into ceca which extend about three-fifths the distance caudad. Paired groups of mucin-gland ducts empty their slimy contents at the outer margin of the oral sucker. Each duct opens thru a hollow piercing spine which caps the duct (Fig. 5). Each group can be traced back to three mucin glands in the region of the acetabulum. These cells have loosely scattered granules in the cytoplasm and large nuclei. No other mucin glands have been found. Several germ cells have been found in the region of the body posterior to the acetabulum. The number is considerably in excess of the number of testes in the adult worm.

In specimens of the cercariae of *Schistosoma mansoni* from Caracas, Venezuela, which the writer has been enabled to examine thru the courtesy of Dr. Juan Iturbe, the mucin glands consist of only two pairs of cells of the granular type, but, in addition, four pairs of a non-granular type, somewhat smaller and surrounding the granular cells. The ducts are decidedly heavier than in the South African species. They open thru six spinose protuberances which cap the ducts.



To clear up Iturbe's work on this species it is necessary to state that the figure in Iturbe's paper (1917) is not a photomicrograph, but rather a diagram more nearly corresponding to the cercaria of *S. japonicum* than to the actual cercaria of *S. mansoni* which Iturbe found in the vicinity of Caracas.

Recently Cort (1919) has made a study of the cercaria of *S. japonicum*, which is altogether the most thorough anatomical analysis yet made of a human schistosome larva. The number of flame cells on each side of the body is four, rather than five as Miyairi and Suzuki found; likewise, the number of mucin gland cells is five for each side of the body, whereas the Japanese investigators considered the number to be three. As he says, the difficulty in differentiating the three human schistosome cercariae "is undoubtedly due to the limitations of our knowledge than to a lack of specific differences."

TABLE FOR DIAGNOSIS OF SPECIES OF HUMAN SCHISTOSOME CERCARIAE

	<i>S. haematobium</i>	<i>S. mansoni</i>	<i>S. japonicum</i>
Size:			
Body	240 x 100 $\mu$	140 x 60 $\mu$	100-210 x 66 $\mu$
Tail trunk	200 x 47 $\mu$	200 x 27 $\mu$	150 x 20 $\mu$
Furci	80-100 $\mu$ long	50 $\mu$ long	75 $\mu$ long
Oral sucker	60 $\mu$ in transection x 64 $\mu$ in length	30-34 $\mu$ in transection x 30-34 $\mu$ in length	33 $\mu$ in transection x 54 $\mu$ in length
Mucin glands	3 pairs with large nuclei and granular acidophilic cytoplasm	2 pairs with large nuclei and granular acidophilic cytoplasm; 4 pairs with small nuclei and basophilic slime contents	5 pairs with large nuclei and granular acidophilic cytoplasm
Mucin ducts	Moderately thick	Very thick	Very thick
Duct openings	At anterior end of oral sucker; capped by 3 pairs of hollow piercing spines	At anterior end of oral sucker; capped by 6 pairs of hollow, piercing spines	At anterior end of oral sucker; capped by 5 pairs of hollow, piercing spines
Germ cells	Several large cells posterior to acetabulum	Many cells at posterior end of body	Clustered mass of cells just behind acetabulum
Parthenita	Sporocyst	Sporocyst	Sporocyst

Neither in Cort's description nor his figures is the exact relationship of the piercing spines to the openings of the mucin gland ducts clear. While he may be correct in assuming that these glands "of the fork-tailed cercariae appear to be homologous to the stylet

glands of the xiphidio-cercariae which open at the base of the stylet or piercing organ, and to which the function of dissolving tissue in connection with the penetration of the cercaria into its host has been ascribed by certain authors," he is not exact in his statement that "instead of a single stylet as in the xiphidio-cercariae, the schistosome cercariae have a number of spines around the openings of the cephalic gland which perform the same function as the stylet in penetration." For each one of these piercing spines, hollowed in the center, caps the opening of a mucin gland duct, and it is thru this hollowed spine that the secretion of the gland is poured forth (Figs. 4, 5). This has been clearly demonstrated not only in the cercariae of *S. haematobium* and *S. mansoni* and in *Cercaria gladii*, but also in many undescribed schistosomes which the writer has studied, as well as the larval echinostome, *C. acanthostoma* Faust. The numbers of piercing spines for each mucin gland group is five in the cercaria of *S. japonicum*, altho Cort has figured five on one side of his drawing, and only four on the other side. This exact relation of a piercing spine to the opening of each duct has been borne out in every case which the writer has examined. Thus for *Cercaria gladii* and the cercaria of *S. haematobium* there are paired groups of three hollowed spines, while in the cercaria of *S. mansoni* there are six piercing spines to each group.

*Cercaria catenata* Cawston 1917 (Fig. 6)

Cawston's basis of diagnosis of this species which was found in *Planorbis pfefferi*, *Lymnaea natalensis* and *Physopsis africana* at Durban, Natal, was the "chain of blackish granules" which "lay on each side of the divided alimentary canal." Even with this apparent distinction Cawston has labelled certain specimens of *Cercaria catenata* "tadpole cercariae." A more thoro study shows the presence of a collar of spines just behind the oral sucker. This fact, among others of critical value, places the species among the larval Echinostomidae.

Contrary to Cawston's designation of the species as a large form (1917: 131), the writer has found it to be small, especially small for the group to which it belongs. The body measures 0.26 mm. in length by 0.13 mm. in width. The tail is 0.4 mm. long and only 36 $\mu$  in diameter at the base. The oral sucker is 24 $\mu$  in section, and the acetabulum 43 $\mu$ . The latter is sunk in a deep circular depression which has a diameter about twice that of the acetabulum.

The oral sucker leads thru a very short prepharynx into a small pharynx. From the pharynx the esophagus, almost capillary in structure, runs posteriad to the anterior margin of the acetabular depression, where it gives rise to ceca of very small diameter. These end just behind the posterior margin of the depression.

The main tubules of the excretory system are characteristically echinostome. From the sides of the transversely compressed bladder two lateral collecting tubules run forward just outside of the ceca. They continue almost as far as the plane of the pharynx where they flex outward and backward. A single median tubule runs backward from the bladder thru the tail. It forks at the very end of that organ so that two outlets are formed. Cawston's "chain of blackish granules" refers to the granules in the excretory system.

The germ cells in the cercaria consist of a group of units in the median line posterior to the caudal margin of the acetabular depression and another mass on the anterior margin of the depression. These are connected by a chain of cells. The body of *Cercaria catenata* is crowded with cystogenous cells. The cyst granules in the cells are long narrow bodies closely packed together side by side. Judging from this provision, encystment must occur rapidly in this species.

Cawston has described the redia of *C. catenata* as a robust animal with four walking legs, a pointed posterior end, and a large gut filled with orange-colored material.

*Cercaria constricta* nov. spec. (Fig. 7)

Liver tissue of *Physopsis africana* which Cawston sent from Natal has been found to contain rediae and cercariae of a new larval echinostome for which the name *Cercaria constricta* is proposed. The body of the larva measures 0.19 mm. in length by 0.1 mm. in width, while the tail averages 0.28 mm. in length by about  $40\mu$  in section at the base. The entire body and tail are covered with very sharp spines directed posteriad. In the region lateral to the pharynx there is a deep constriction of the body which sets off the head from the trunk. A collar of sharp spines, larger than those covering the body as a whole, runs in a single series along the anterior margin of the constriction. It is complete except for a small gap on the ventral side just below the pharynx.

The acetabulum has a width measurement of  $53\mu$  and a length diameter of about  $35\mu$ . The oral sucker has an average diameter of about  $35\mu$ . The digestive tract consists of a short prepharynx, a small muscular pharynx, a long esophagus reaching to the anterior margin of the acetabulum, and furci which extend to the subcaudal region of the body. Mucin glands have been seen, but their arrangement in the body has not been exactly determined.

The excretory bladder is quadrangular in shape and quite muscular. From the median posterior margin a single caudal tubule runs half the distance distad, where it splits into two tubules. These tubules open laterad in the subdistal region of the tail. An anterior collecting

tubule from each of the anterior angles of the bladder runs forward to the plane of the pharynx, where it reflexes and continues backward. Further than that the writer has not been able to make out its course. The tubules contain no excretory granules.

Conspicuous germ-gland cells have been observed just anterior to the acetabulum.

The redia is an elongate sac without feet, with a minute obovate pharynx, a simple gut and a deep constriction in the region of the neck. A birthpore has not been observed. The redia is entirely covered with spines.

It will be noted that *Cercaria constricta* differs from *C. catenata* in several important points. Prominent among these are the spinosity of the cercaria and redia of *C. constricta* and lack of such integumentary differentiation in *C. catenata*; difference in type of excretory tubule in tail, and absence in *C. constricta* of the circular depression found around the acetabulum of *C. catenata*. The lack of excretory granules in *C. constricta* is probably of specific value, but this point needs checking with living material. A further distinction between the two species is found in the presence of four feet in the redia of *C. catenata*, while the redia of *C. constricta* lacks feet entirely.

That *Cercaria arcuata*, described by Cawston (1918a:95) from the Transvaal, is still a different species of larval echinostome is shown from the fact that the species has two redial feet, and an anterior collecting system which empties thru a single median stem into the bladder.

*Cercaria arcuata* Cawston 1918 (Fig. 8)

The original description of this species was made by Cawston on the material from *Isidora* sp. from the Schoonpoort at Klerksdorp; the material which the writer has had an opportunity to examine is from *Lymnaea natalensis* from Natal. As in other echinostome larvae which Cawston has described, he has referred to this species as a "leptocercous cercaria" (1918a:95).

The body of *Cercaria arcuata* is 0.15 mm. long by 0.1 mm. wide in the region of the acetabulum. The tail is about one and one-half the length of the body. The body is entirely covered with spines which are most conspicuous at the anterior end. A distinct collar prominence has been seen on the worm, but no collar spines have been made out with certainty. The anterior end of the body is capable of considerable extension. The oral sucker measures  $37\mu$  in diameter, and the acetabulum has a diameter of  $33\mu$ .

The redia is a long sac with a pair of pointed feet about in the middle of the body. At the anterior end of the body pharynx, collar prominence and birth pore can be readily seen. The rhabdocoel gut



may be small or large, depending on the age of the redia and the amount of food ingested.

Contrary to Cawston's description, the writer has been able to make out a prominent pharynx just dorsal to the oral sucker. It leads into a long esophagus. The furci arise just anterior to the acetabulum and continue nearly to the posterior limit of the body.

The excretory bladder is an elongate median organ extending nearly to the posterior margin of the acetabulum. Here it forks in horse-shoe fashion to form two dilated collecting tubules which run forward to the region of the pharynx. It is filled with a few very large excretory granules, the "chain of cystogenous vesicles" of Cawston. The finer portions of the collecting tubules have not been made out. The collecting tubule in the tail forks soon after it enters that organ. The two branches continue distad and open to the exterior in the subdistal region of the organ.

The body is filled with an enormous amount of cystogenous granules, which obscure all the finer structure of the worm. On encystment these granules are extruded from the body, forming a covering around the decaudated worm.

*Cercaria cawstoni* nov. spec. (Fig. 9)

This is one of the "tadpole cercariae" which Cawston has recorded for *Physopsis africana* and *Lymnaea natalensis* from Natal. A comparison of the figure accompanying this description with Cawston's several figures of this type shows how entirely inadequate and misleading his description is.

The larva is ovate-oblong, with a slight protrusion at the oral end and an impocketing at the posterior end into which the tail fits. The body measures 0.38 mm. in length by 0.21 mm. in width. The tail measures 0.31 mm. in length by  $43\mu$  diameter at the base. The body is entirely covered with minute spines, but the tail is aspinose. At each side of the caudal pocket is a studded cluster of long, heavy spines, which are imbedded in a thickened region of the integument (Fig. 9, 9 b). The oral sucker has a diameter of  $43\mu$ , while the acetabulum measures about  $60\mu$ . Imbedded in the dorsal wall of the oral sucker is a stylet,  $27\mu$  long. This organ is of the simple quill type, but is unique in having a median longitudinal reinforcement in addition to the usual transverse thickening (Fig. 9 a).

From the oral sucker the digestive track is traced thru a short prepharynx to a minute pharynx,  $16\mu$  in diameter. From this region a short esophagus leads to the ceca which extend nearly to the posterior end of the body. Emptying at the sides of the stylet are paired groups of mucin gland ducts, each group arising from four mucin

cells. These cells have large vacuolated nuclei and a homogeneous chromophilic cytoplasm. The excretory bladder is a transversely constricted organ into which empties a common collecting tubule. This tubule forks just posterior to the acetabulum. A single mass of germ cells has been found posterior to the acetabulum.

The body of *Cercaria cawstoni* is filled with cystogenous granules. When the cercaria is freed from the ruptured sporocyst it drops its tail and encysts.

*Cercaria frondosa* Cawston 1918 (Fig. 10)

*Cercaria frondosa* is a sturdy amphistome larva measuring 0.4 mm. in length by 0.31 mm. in width, with a tail 0.43 mm. long by  $57\mu$  in section at the base. The oral sucker has an average diameter of  $66\mu$ , while the acetabulum,  $95\mu$  in diameter, is situated at the posterior margin of the body and not on the ventral side as Cawston has figured it (1918: 69). The parthenita is a large, heavily walled muscular redia, varying in size, but always possessing a prominent pharynx, a long, slender gut and a birth pore. The worm was found in *Isidora schakoi* at Potchefstroom, Transvaal.

Internally, the oral sucker leads into a cavity with distinct pharyngeal pockets, which probably argues for its place among the Diplodiscinae. The short esophagus is not surrounded by a postpharyngeal sphincter. It opens into a rather inconspicuous pair of ceca which run posteriad to the region of the acetabulum. The bladder is small, but collecting tubules which empty into it from the side, are enormously dilated by excretory granules. The two main tubules can be traced forward to a region under the pigment areas immediately behind the eye-spots. A single tube, running thru the middle of the tail, forks near the proximal end of that organ to open thru small pores to the exterior. The germ cells of the cercaria consist of a clump of elements in the median line just behind the anterior limit of the lateral excretory tubules.

Two eye-spots are present. They are of a bee-hive shape, with the pigment cup opening anterolaterad. The optic cell is conspicuous in the young cercaria long before the pigment granules accumulate around it. Spreading out from the eyes in frondose arrangement are pigment elements which show under high magnification a grouping into flaky masses that at times extend over the entire dorsal surface of the animal.

Large rhabditiform cystogenous granules pack the parenchyma cells of the cercaria. Upon the maturing of the cercaria either within the liver gland of the snail or after wandering out of the host, the cystogenous granules are thrown out to form a heavy cyst membrane

With encystment the tail is dropped and the larva passively awaits transmission to the subsequent host. Cawston has called this larva a leptocercous distome.

*Cercaria fulvoculata* Cawston 1919 (Fig. 11)

Cawston called this species leptocercous, but it must be placed among the larval monostomes. It is ovate in outline, with slight auricular prominences on each side near the anterior end. The body is 0.4 mm. long and half as wide. The tail is heavy and about 0.6 mm. long. It is provided with six paired groups of long falciform cells surrounding the caudal excretory canal. The body has a small but prominent oral sucker and a pair of aspinose caudal pockets. The cercaria is binoculate, with flecks of pigment surrounding the eye spots and at times extending backward along the nerve tracts. The cercaria was taken from *Lymnaea natalensis* at Durban.

A large bladder lies mesad near the posterior end of the body, with lateral conduits opening into it from the sides. These ducts connect with one another just posterior to the eyes. In front of the bladder is an ovarian cell mass, and ventral to the cornua of the bladder are small testicular germ masses. Ducts from these glands run forward in parallel courses to the region of the genital pore, which is situated behind the plane of the eye spots.

The redia is a simple sacculate structure with medium-sized pharynx and long slim gut. There are no feet. A birth pore has not been seen.

#### DISCUSSION

A survey of the data above shows the inadequacy of Cawston's descriptions. In a private communication he has stated that the illustrations are most unsatisfactory, but are very similar to those of Drs. Leiper and Atkinson in the *British Medical Journal*. This is no less unfortunate. It is true that Leiper's descriptions and figures will not serve to separate species of larval schistosomes, because all details of structure are omitted. Leiper's statement (1915: 39) that the systematic position of a bifid-tailed cercaria can only be effectively established in the first instance by experimental infection of a susceptible host and the subsequent examination of the adult resulting therefrom is misleading and entirely out of accord with the facts. Specificity of structure is as characteristic of the larval fluke as of the adult, and failure to find specific differences between cercariae is due to inadequate observation.

These difficulties are generally felt, and Leiper (1918: 168) states that there is no evidence that the various forms so loosely and repeatedly termed "*Bilharzia cercariae*" in Cawston's numerous papers

## EXPLANATION OF PLATE

Fig. 1.—*Cercaria gladii*, ventral view, showing digestive, nervous and reproductive systems.  $\times 170$ .

Fig. 2.—*Cercaria parvocolata*, ventral view of body only, showing digestive and germ-cell glands.  $\times 330$ .

Fig. 3.—*Cercaria* of *Schistosoma haematobium*, ventral view, showing digestive glands and germ cells.  $\times 170$ .

Figs. 4 and 5.—Anterior tip of cercaria of *Schistosoma mansoni*. 4, mucin ducts and openings at anterior margin of sucker;  $\times 330$ . 5, tips of ducts, showing hollow spine capping each duct.  $\times 990$ .

Fig. 6.—*Cercaria catenata*, ventral view, showing digestive and excretory systems and germ cells.  $\times 170$ .

Fig. 7.—*Cercaria constricta*, ventral view, showing digestive and excretory systems.  $\times 170$ .

Fig. 8.—*Cercaria arcuata*, ventral view, showing digestive and excretory systems.  $\times 170$ .

Fig. 9.—*Cercaria cawstoni*, showing digestive and excretory systems and single mass of germ cells; *a*, stylet, enlarged; *b*, lateral view of cluster of caudal pocket spines, enlarged.  $\times 170$ .

Fig. 10.—*Cercaria frondosa*, ventral view, showing pigmentation around eye-spots, digestive and excretory systems and single mass of germ cells.  $\times 170$ .

Fig. 11.—*Cercaria fulvoculata*, ventral view, showing pigmentation around eye-spots, excretory and reproductive systems.  $\times 170$ .



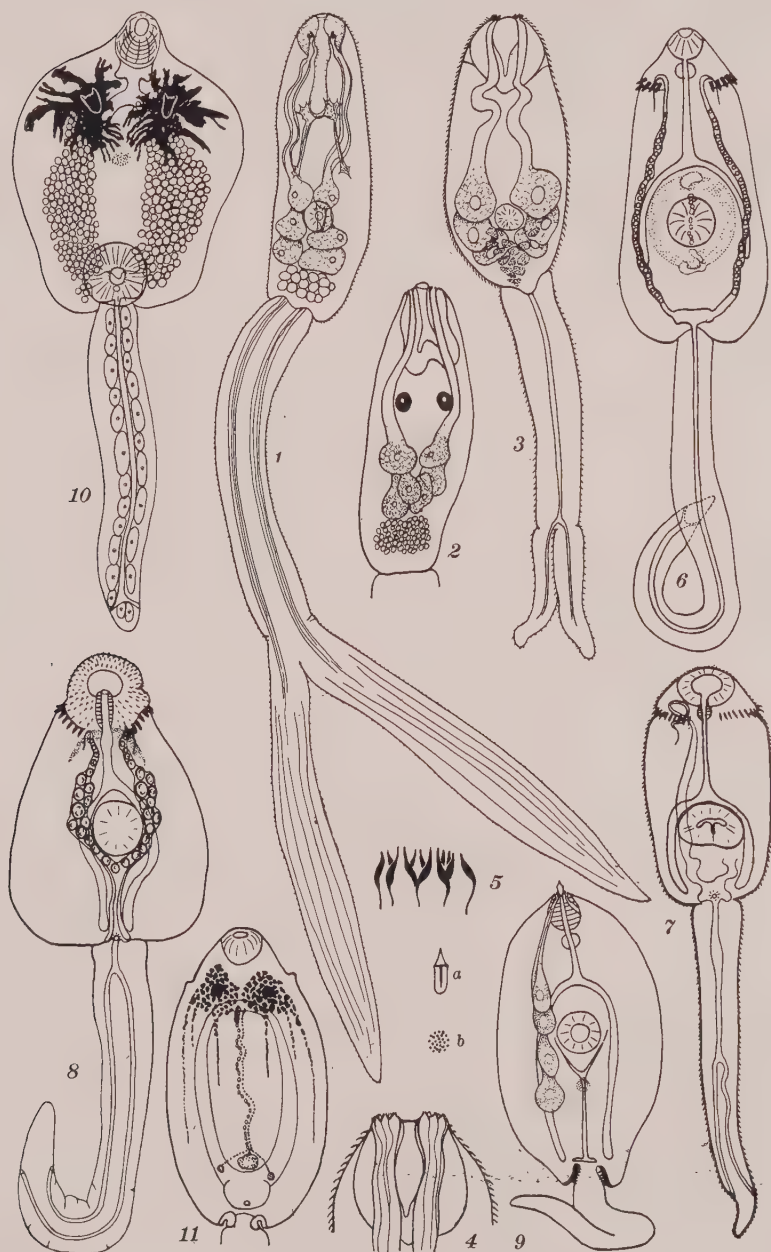


PLATE XVIII



are actually such. In a review of one of Cawston's papers (1916: 348) new figures are substituted for those in the original article (1915: 258). A comparison fails to show a single common likeness between any two of the figures, altho they are labelled "bilharzia cercariae." In another paper Cawston (1916a: 201) has labelled at least three distinct species (Cercaria of *S. haematobium*, *C. oculata* and *C. catenata*) as "human forms of Cercariae," altho the latter forms cannot possibly be considered human forms.

## SUMMARY

1. Ten species of cercariae from South Africa, including two new, are described.

2. The cercariae of *Schistosoma haematobium*, *S. mansoni* and *S. japonicum* are easily distinguished on the basis of number and type of mucin glands and ducts, and their outlets. Differences in arrangement of the germ cells may also be used in this diagnosis.

3. Diagnosis of a larval trematode requires exact data on the size and shape, on the integument, on the excretory system, on the digestive system and on the germ cells.

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*TETRADONEMA PLICANS* NOV. GEN. ET SPEC.,  
representing a new family, TETRADONEMATIDAE as now

found parasitic

IN LARVAE OF THE MIDGE-INSECT *SCIARA COPROPHILA* LINTNER

N. A. COBB

United States Department of Agriculture

HABITAT AND OCCURRENCE

*Number, Location and Maturity of the Parasites.*—Of this parasitic nema, both male and female are found in about equal numbers in the body cavity of the larvae of the midge insect identified by Professor H. B. Hungerford as *Sciara coprophila* Lintner, often as many as six to twelve of the parasites being found in a single larva. Adult males, about one-sixth as long as their mates, are usually found coiled about females, and both males and females are more or less entangled with the malpighian and tracheal vessels of the host, so as often to be rather difficult of extraction. These facts give rise to the specific name *plicans*. The generic name was suggested by the highly interesting four-celled organ, the tetrad, located in the anterior part of the nema. The parasites occupy a very considerable part of the body cavity of the host. In the material examined some of the nemas of each sex were surrounded by cast-off cuticula, indicating that they moult at least once after they enter the host. The fully matured females contain thousands of eggs having somewhat the form of an immature mushroom cap (Fig. 1). The shells of the eggs are smooth and of medium thickness, and contain embryos in various stages of development. The most advanced embryos seen appeared to be taking on a serpentine form and to be coiled once to twice in the egg. When, in the course of dissection of preserved material, the largest females become broken, their eggs escape in large numbers into the surrounding fluid. Preserved eggs escaping in this way are about  $33\mu$  in greatest diameter.

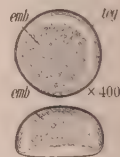


Fig. 1.—Two views of eggs from the uterus of *T. plicans*. emb, embryo; teg, shell of the egg.

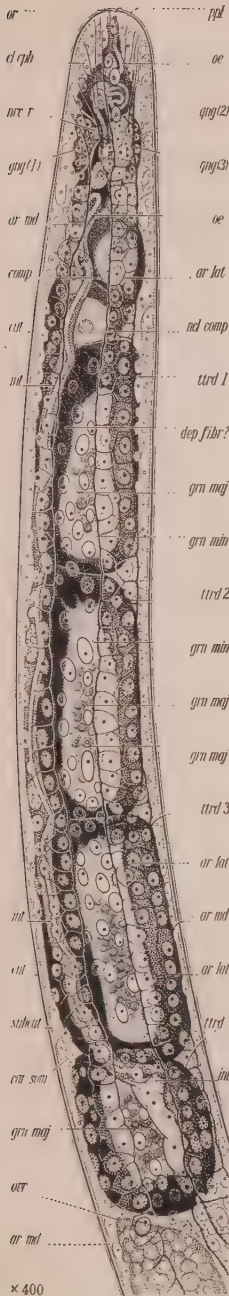
SIZE AND FORM OF TETRADONEMA PLICANS

*Dimensions. The Formula.*—Below are measurements of male and

0.0	0.9	2.2(?)	$\frac{50 \times 60^{140}}{2.4}$	08.5	0.0	5.7	9.3(?)	$\frac{71 \times M}{5.2}$	87.
0.0	0.8	0.8		0.9		3.1	3.6		4.2

female. The figures are averages derived from four males and two females, prepared by fixing the host in picro-aceto-subliminate and pre-





serving in 70 per cent. alcohol, and finally mounting in glycerin jelly. The tissues of the head of *T. plicans* appear susceptible of contraction, and it seems not unlikely, from the appearance of the preserved specimens here described, that the heads had become fixed in a somewhat retracted attitude, and, if so, the measurements must be interpreted accordingly; see Figure 2, in which the tortuous esophagus may indicate a retracted condition of the head. Similar retractions may occur in the heads of other nemas, e. g., *Oxyuris vermicularis*, a fact that has led some writers into a *non sequitur*, and consequent disparagement of the decimal formula. Such changeableness of form has no more to do with the method of expressing the measure-

Fig. 2.—Head end of a mature female of *Tetradonema plicans*, nearly lateral view. The entire tetrad is shown—occupying the greater part of the illustration. *ar lat*, lateral field; *ar med*, median field; *cav som*, somatic cavity; *cl cph*, nucleus of cephalic cell; *comp*, cells referred to as companion cells of the tetrad; *cut*, cuticula; *dep fibr?*, semifibrous deposit between the nuclear membrane and the cell wall of one of the members of the tetrad; *gng* 1, 2 and 3, groups of nerve cells connected with the nerve-ring; *gng maj*, the major granules of the tetrad elements; *gng min*, minor granules of the tetrad elements; *int*, alimentary canal; *nrv comp*, nucleus of cell referred to as companion cell of the tetrad; *nrv r*, nerve ring; *oe*, esophagus; *or*, mouth; *ovr*, ovary; *pbl*, cephalic papillae; *subcut*, subcuticula; *trrd* 1, 2, 3 and 4, the four elements of the tetrad; compare with Figure 3.

ments than the market value of wheat has to do with the currency in which it is expressed. The same may be said of other variabilities of form, as well as uncertainties of observation. The limits of variability may be expressed in the formula in the usual way, by using the two limiting figures. Uncertainty as to measurement may be expressed by an interrogation mark, as in the above formulae, where the location of the beginning of the intestine is indicated as questionable. Notwithstanding the uncertainty, the queried figures,  $\frac{9.3}{3.6}$  and  $\frac{2.2}{0.8}$ , are as useful as ever in indicating the contour of the body.

#### STRUCTURE OF THE NEMA

*Cephalic Organs and Body-Wall.*—The general appearance of the tissues of the head and of the neck of *Tetradonema* is reminiscent



somatic cells, presumably muscle cells. These narrow and weak longitudinal muscular fields are submedian in position, and each is represented by about four striae. The transparent tissues of the neck permit of seeing these submedian, narrow, longitudinally striated elements more clearly than they can be seen elsewhere, but they have been followed far backward, and no doubt exist throughout the length of the body (*msc*, Fig. 3). There exist in the neck, as elsewhere, not only lateral fields, but also median fields. In one of these, the ventral, the row of nuclei is more definite and the nuclei are somewhat larger than in the other.

#### DEGENERATE ALIMENTARY CANAL

The digestive system of the adults of both sexes is more or less degenerate or vestigial, as, it appears, is often the case with nemas inhabiting the body cavities of insects, e. g., *Allantonema*, *Sphaerularia*, *Tylenchus*, etc. It may not be an unreasonable supposition that to some extent the food of *T. plicans* is absorbed through its cuticula, since fully adequate means for imbibing it through the mouth seem to be lacking.

*Alimentary Canal, Male.*—There is no pharynx. From the mouth opening backward the esophageal tube is very narrow, but may present an almost imperceptible swelling just in front of the nerve-ring. This latter lies about half way to the tetrad and is nearly transverse. There are filamentous processes passing from it to the body wall, presumably nerves. Near the front of the tetrad may be dimly seen what appears to be the junction of the esophagus with the intestine. Just in front of this point the esophagus is very slightly swollen; the posterior portion of the esophagus therefore appears somewhat narrowly clavate in form, and is about one-fourth as wide as the corresponding part of the neck. In some specimens, however, this swelling was sought in vain. The succeeding part of the alimentary canal (the intestine, or the posterior part of the esophagus, as the case may be) is at first about as wide as the part of the esophagus just described, but soon diminishes in size and becomes a rather insignificant looking strand of indefinite tissue, containing a faint lumen. The alimentary canal soon passes to the ventral part of the body, and is so inconspicuous and deteriorated that one is usually unable to follow it further. In one case, at the nerve ring, which was about half way back to the first member of the tetrad, the esophagus was not more than one-fifth as wide as the corresponding part of the neck, and thence backward it diminished in size and was very difficult to follow. In the male the esophageal lumen does not seem to lead through a granular plasma as is the case in the female illustrated in figures 2 and 4. In immature males a rectum is present, and, joining it, a portion of the

intestine can be seen, extending forward a distance somewhat greater than the length of the tail, and at its widest part becoming half as wide as the body. At a point as far in front of the anus as the terminus is behind it, the intestine is smaller, and farther forward still it is difficult to follow and seems very rudimentary. This condition of things exists in those males whose testes are filled with spermatocytes about one-fifth as wide as the body; that is to say, somewhat immature males.

*Alimentary Canal, Female.*—An examination of the females shows that, just as in the male, the alimentary canal is much deteriorated, but the details are somewhat different (Fig. 2, *int*). Immediately behind the mouth opening the lumen of the canal becomes tubular and more or less tortuous. The diameter of the more or less corrugated lumen is about equal to the thickness of the cuticula. Surrounding the median canal is a granular tissue or "plasma" in which large nuclei are to be seen here and there. From the mouth backward this granular tissue expands so that at a distance from the anterior extremity one and one-half times as great as the diameter of the head it may become about half as wide as the corresponding part of the neck. Immediately behind this point, however, there is a constriction, and in the midst of this constriction the nerve-ring is found (Fig. 2, *nr v r*). Immediately behind the nerve-ring the intestinal canal, or esophagus, as I believe we may still term it at this point, gradually widens until it becomes one-third as wide as the body; it then again diminishes in size so that anterior to the two large cells in front of the tetrad it is only about one-fifth as wide as the corresponding portion of the body. This portion of the body seems to be what would be called the base of the neck, and if so, this constriction, for such it seems to be, corresponds with the beginning of the intestine. Behind this constriction the alimentary canal again widens and soon becomes about one-fourth as wide as the body, and then once more begins to decrease. However, even as far back as the last member of the tetrad it still appears to have a tubular lumen. At this point the tubular lumen suddenly ceases, suggesting the possibility that the esophagus really extends farther back than indicated above (Fig. 2, *int*). No trace of the alimentary canal was seen farther back than the middle of the body. As the alimentary canal, for a certain distance at least, has a distinct lumen, and there is a distinct mouth opening, small though it be, it seems likely that the intestine is still capable of taking in liquid nutriment. It is possible that the large nuclei associated with the anterior part of the alimentary canal, of which half a dozen may be counted in the esophageal portion just described, may have something to do with assimilation. It has been assumed that certain nemas parasitic in the



body cavities of insects absorb their nutriment through the cuticula, and there is good cause to suppose that in some instances this may be

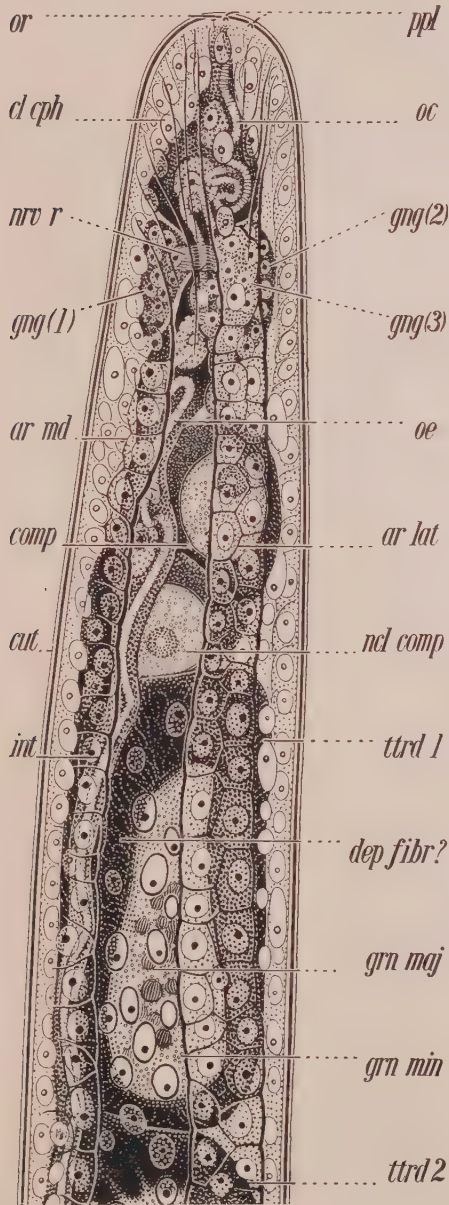


Fig. 4.—Head end of a mature female of *Tetradonema plicans*; nearly lateral view. Compare with Figure 2. *ar lat*, lateral field; *ar med*, median field; *cl cph*, nucleus of cephalic cell; *comp*, cells referred to as companion cells of the tetrad; *cut*, cuticula; *dep fibr?*, semifibrous deposit between the nuclear membrane and the cell wall of the front member of the tetrad; *gng* 1, 2 and 3, groups of nerve cells connected with the nerve ring; *grn maj*, major granules of the tetrad; *grn min*, minor granules of the tetrad; *int*, alimentary canal; *ncl comp*, nuclei of one of the cells referred to as companion cells of the tetrad; *nrv r*, nerve ring; *oe*, esophagus; *or*, mouth; *ppl*, cephalic papillae; *ttrd*, one of the elements of the tetrad.

#### THE TETRAD

*Structure of the Tetrad.*—A very striking feature of the anatomy is the occurrence, toward the head end, of four large unicellular organs, arranged tandem, and occupying in this region the greater portion of the body cavity (Fig. 2, *ttrd*, 1, 2, 3 and 4). It is this quartet of bodies, the tetrad, that gives rise to the generic name *Tetradonema*.

That each of the four elements of the tetrad is unicellular is shown by the fact that in the younger nemas, where the tetrad is smaller, it is

quite clear that each of its elements is a single cell containing a large spherical nucleus having a distinct nucleolus. In the full grown female the tetrad is much more strongly developed, and the entire space between the cell wall and the nuclear membrane of each element is completely filled with a semi-fibrous, semi-granular deposit (Figs. 2 and 4, *dep fibr?*). Under these circumstances the form of the cells is no longer spherical, but more or less cylindrical, owing to very great increase in size and consequent pressure from surrounding organs. In the males, also, the tetrad cells may become so large that the nuclei are no longer spherical, though this seems less commonly the case than in the females. The nuclei of the tetrad may become half as wide as the body, the deposit surrounding them then becoming thicker and more opaque, and seeming to be more "fibrous" the older it becomes. The tetrad in the male may be of such a size as to be twice as long as the distance between its anterior extremity and the mouth opening. Accompanying the tetrad, and in front of it, are to be seen two smaller more or less spheroidal cells that seem to be larger in the female than in the male (Figs. 2 and 4, *comp*). Occasionally each of this pair of cells is so large as to suggest that they are "companions" of the cells comprising the tetrad.

*Function of the Tetrad.*—Such a striking organ as the tetrad of *Tetradonema* cannot but give rise to the question, "What is it for?" I have been unable in the examination of the small amount of preserved material available to make out the histological connections of the tetrad, but the following facts are clear as a result of the examination made.

1. The organ is found in both sexes in the same form and consists of four unicellular, apparently equivalent, components, which develop from comparatively normal cells lying near the base of the neck, the usual location of the renette, and seem to grow with the age of the organism rather than with its size.

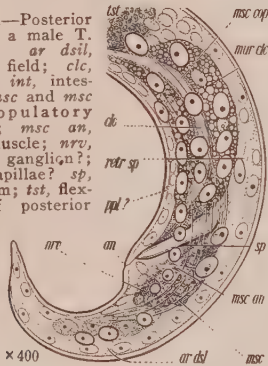
2. As the nema ages these elements not only increase in size, but also change in structure, one of the principal changes being a bulky, apparently semi-fibrous deposit just inside the cell wall. Meanwhile the nucleolus becomes a locus rather than a cell organ, and finally there are deposited in the very much enlarged nucleus, relatively large, spherical, more or less structureless granules (Figs. 2 and 4, *grn maj*). Sometimes these major granules have a distinctly refractive element. As these larger granules appear only in the larger tetrads, the possibility is suggested that they are a degeneration phenomenon, but I am more inclined to regard them as excretory in nature, along with the semi-fibrous matter outside the nucleus. The nucleus maintains its membrane to near the close of the history of the tetrad.

3. It is noticeable that no trace of the usual excretory pore and renette has been seen in any of the specimens.

Is it possible that the cells of the tetrad are storehouses for excreta? The food of this nema may be predigested, but the catabolism must give

rise to waste matter. Now there is no true anus; nor has any excretory pore been seen. To this statement of the entire absence of the main channels through which excreta are usually voided, may be added the suggestion that should the parasite pour its excreta into the body fluid of the host, presumably the effect on the host would be injurious, and this in turn would be inimical to the parasite. If such a thing were possible, it would seem advantageous to the nema

Fig. 5.—Posterior end of a male *T. plicans*. *ar dsil*, dorsal field; *clc*, cloaca; *int*, intestine; *msc* and *msc cop*, copulatory muscle; *msc an*, anal muscle; *nrv*, anal ganglion?; *ppl*, papillae? *sp*, spiculum; *tst*, flexure of posterior testis.



under the circumstances to store up within its body the wastes of its own metabolism, the excreta due to its growth and reproduction. The data thus far disclosed leave it possible to suppose the tetrad to have some such function. On the other hand, no such organ is known in any other parasitic nema, though of course it is conceivable that organs having the function here imagined but more obscure or of smaller size, might hitherto have escaped notice. It is desirable that the tetrad be studied in living specimens, and be submitted to chemical tests.

*Sexual Organs of the Male.*—The single spiculum is median in position, and without accessories. There is no bursa and there are no ventral supplementary organs, and no distinct caudal papillae. Oblique copulatory muscles are to be seen for some distance in front of the anus. The ejaculatory duct is about one-half as wide as the body. In somewhat immature males the main portion of the two testes already nearly fill the body cavity, and contain many thousands of spermatocytes, whose average diameter is about one-fifth to one-sixth that of the body. Immediately behind the tetrad a flexure is to be seen in the anterior testis. At this point the testis suddenly diminishes considerably in size and extends thence backward and ends; this blind end of the anterior testis seems to lie toward the middle of the nema, and is nearly one third as wide as the corresponding portion of the body. Toward the posterior end of the nema, as far in front of the anus as the terminus is behind it, there is a definite

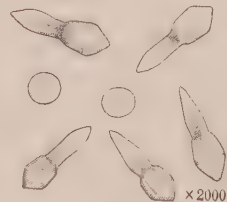


Fig. 6.—Sketches of the sperm cells of *Tetradonema plicans*. From various points of view.

broad contour line indicating the presence of a flexure of a similar character in the posterior testis. The two testes meet near the middle of the body where their junction with the vas deferens is more or less plainly visible. The fully mature spermatozoa found in the *vas deferens* of the male and in the uterus of the female are somewhat asymmetrically elongated-fusiform bodies about one-sixth as long as the body is wide, and about one-third as wide as long. Packed together with them in the proximal portion of each testis are more or less finely granular, spheroidal, apparently non-nucleated bodies of various sizes, the largest being one-fourth as wide as the body of the nema, and the smaller having not more than half this diameter. As these spheroidal bodies are most numerous and most apparent near the proximal ends of the testes, it is assumed that in the ripening of the sperm these bodies are formed. The ripened spermatozoa occur in thousands and are reminiscent of those of *Mononchus*, *Dorylaimus* and *Anticoma*. In the uteri of the female they have more or less the contour of tadpoles (Fig. 6). The sperm cells appear to be produced in groups, possibly in groups of four. It was found difficult to make an accurate count, so closely were they packed in the testes, but the number of individuals in a group is certainly small.



Fig. 8.—Tail end of female. *ar ds*, lat and *vnt*, the dorsal, lateral and ventral fields; *cl* or *lat*, *vnt* and *ds*, cells of the fields; *ovr*, posterior flexure of the ovary; *str*, striae.



Fig. 7.—Optical section vulvar region of *T. phicans*. *ar vnt*, ventral field; *cav som*, somatic cavity; *cav*, cavity of the vagina; *cut*, cuticula; *ds ut*, uterine duct; *ov im*, oocyte; *subcut*, subcuticula; *ut*, uterus; *vag*, wall of the vagina; *vlu*, location of the vulva.

*Sexual Organs of the Female.*—From the rather inconspicuous vulva, the very strongly developed vagina passes inward at right angles to the ventral surface three-fourths the distance across the body. It is composed of a bulky mass of cellular tissue three-fourths as wide as the nema, and is one of the main features of the middle of the body (Fig. 6). Its distinct cavity appears rather narrow, and is lined with a layer consisting of scores of closely packed, relatively large, elongated cells. This more or less columnar “epithelial” lining is the main feature of the developing vagina. Outside it, however, there is a layer of smaller cells placed somewhat irregularly. From the proximal part of the vagina, toward the dorsal side of the body, two comparatively narrow tubes lead in opposite directions to the two uteri, one in front, the other behind. Each uterus is about

three to four times as long as the body is wide, and in young females appears to be about half as wide as the body. Where the uterus joins



the ovary there is a faint constriction, and the contour of the organs is here so definite as to make it evident that the ovaries are reflexed (although I usually found it impossible to clearly identify the reflexed portion throughout its length), for a longitudinal optical section at this portion of the body discloses ovarian tissue other than that comprised in the portion of the ovarian tube that joins the uterus. This ovarian tissue extends to near the vagina, from which it is evident that the reflexed portions of the ovaries reach back to near the vulva. These details were made out from the study of nearly mature but unfertilized females. The flexure in the anterior ovary is at the back end of the tetrad; the flexure in the posterior ovary is only a short distance from the end of the tail (Figs. 2 and 8 *ovr*).

*Tail*.—The female has no anus, nor is there any vestige of such an opening. However, its former location may be estimated from consideration of the position of the anus on the male, whose tail end has a similar form. With this estimate in mind it becomes evident that the tail of the female begins to taper some distance in front of the position of the theoretical anus. The tail of the female is rather like that of the male in form, being at first conoid, but ending in a subcylindroid or somewhat convex-conoid terminus, about one fifteenth as wide as the body, and about two to three times as long as wide, and having a more or less acute tip. Considering the tail to comprise that portion of the posterior end extending from the flexure of the posterior ovary to the terminus, this final narrow portion of the tail occupies about one-third of its length. On this portion of the tail the transverse striae of the cuticle can be seen more plainly than on almost any other part of the body. There are, of course, no caudal glands.

*New Family of Nemas*.—I consider *Tetradonema*, of which the type species is *Tetradonema plicans*, to be the type genus of a new family, the Tetradonematidae.

TETRADONEMATIDAE fam. nov.

TETRADONEMA gen. nov.

Small naked insect-parasites with minute males; cuticula wingless, minutely transversely striated; head rounded, tail acute; mouth minute, lipless, oral papillae minute, anus none; esophagus simple, with lumen, intestine vestigial; male and female gonads double, symmetrically reflexed; vulva central, uterine eggs numerous, asymmetrical, containing embryos; spiculum single without accessories, supplements and bursa none.

BIOLOGICAL NOTES ON *TETRADONEMA PLICANS*,  
COBB, A NEMATODE PARASITE OF *SCIARA*  
*COPROPHILA* LINTNER

H. B. HUNGERFORD  
University of Kansas

INTRODUCTION

In January, 1915, while studying the life history of the Mycetophilid fly *Sciara coprophila* Lint., which is often found breeding in potted plants, the writer found one batch of maggots parasitized by a peculiar nematode. The unusual appearance of this worm led to a study of its life history. Several photographs and figures were made at the time as a matter of interest, but aside from recording the effectiveness of this parasite in the destruction of *Sciara* maggots,\* nothing was done until the beginning of 1918, when the nema under discussion was sent to Dr. N. A. Cobb for determination.

DISTRIBUTION

In an endeavor to determine the range of distribution, the number of specific hosts and the percentage of infestation in nature, a careful search has been made of every *Sciara* fly and maggot obtainable. These were collected from fields of alfalfa and of wheat, from meadow grasses, from beneath the leaves in the woods, conservatories, and from green houses, but aside from the material cited at the beginning of this paper, none was ever found parasitized by this nematode. Grub worms and angle worms living in similar situations have been found free also. Experiments to infect these last have failed, although I have seen small angle worms swallow the eggs on several occasions. These angle worms were, in every case, isolated, and kept under observation. They did not become infected.

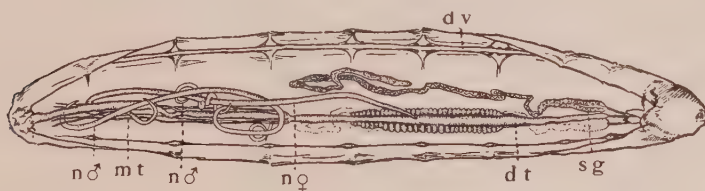
A study of the one lot of infected *Sciaras* and those artificially infected in the experimental work shows that the gravid female parasite may be found in larvae, pupae and adult flies. From two to twenty parasites of both sexes may be found in a single host; on the average, ten worms came from each host, and the number of males ran a little greater than of females.

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\*In the Journal of Economic Entomology for December, 1916, the writer mentioned this nematode as an enemy of *Sciara* maggots and figured the gravid female nematode.

## APPEARANCE OF AFFECTED HOST

When one compares a normal maggot with an infected one, a marked contrast is noted (Figs. 1 and 2). In the former, the maggot appears white, due to the large definite fat bodies present. There are present also segmentally arranged fat masses about the spiracles. The head capsule is shiny black, and, as shown, not conspicuously smaller than the diameter of the anterior part of the maggot. On the other hand, the parasitized maggot gives no evidence of either the long fat masses or the segmentally arranged fat bodies. The head capsule is very likely to be small, indicating the failure of the maggot to make its normal moults. The nematodes within appear white by reflected light (Fig. 2). An examination of such a maggot shows plainly the pulsations of the dorsal vessel, with its muscular wings and pericardial cells, are most plainly seen. In one maggot the female parasite was disrupted in such a way as to release her eggs into the body cavity of the maggot. These eggs were seen buffeted about within the transparent skin of the maggot by the movement of its organs. The dorsal



*Sciara* maggot dissected, *d v*, dorsal vessel; *d t*, digestive tract; *s g*, salivary gland; *m t*, malpighian tubules; *n*, nematode.

vessel was burdened with the continuous coursing of nematode eggs from the rear to the front. Several times the eggs temporarily clogged the dorsal vessel near its anterior outlet, only to break away under the pressure like a released log jam and go racing on with the circulatory fluids.

## EFFECT UPON THE HOST

*Larva*.—The young maggot of 2 mm. may have the body teeming with newly hatched nemas, which are visible under the microscope, without showing much deviation from normal, but very shortly the long fat bodies begin to decrease in size. In the normal maggot these fat bodies are long masses that cling to the salivary glands and surround the malpighian tubules, and the lateral masses, segmentally arranged close to the muscles of the body, one on either side, also a smaller mass beneath each nerve ganglion. These finally disappear altogether, leaving the body very clear. The digestive tract, with its appendages, the heart and the nervous system, can all be seen plainly.

A little later the digestive tract seems subnormal in diameter, but still functional. If the maggot is slit longitudinally, fixed, stained, and the muscular portion and integument spread and mounted upon a slide, the muscular system will present a conspicuous appearance, all the nuclei staining well. The maggot at this stage retains muscular activity, but later remains extended and incapable of movement. A stained mount of such a maggot would show the muscular system in the state of collapse, the heart action being about the only evidence of life. In spite of the general disintegration, the imaginal disks are present, as also the lateral segmentally arranged oenocyte clumps. Maggots, when heavily parasitized, die and disintegrate, leaving within or upon the earth only a mass of several thousand nematode eggs.

*Pupa*.—Sometimes in light or late infection, the maggot endeavors to pupate; it spins out threads and sheets of silk, then contracts, as is usual before transforming. This marks the end of some; others succeed in casting off the larval skin. Many such pupae are little else than mere shells, the body cavity being filled with the egg-burdened nematodes. In no case could I find traces of reproductive organs, either free or attached in infested pupae of either sex.

*Adult*.—Adults emerging from the jars containing infected maggots were in most cases parasitized. They could fly about, but were lacking in reproductive organs. The few uninfected flies contained normal reproductive organs, but there was little difference in the appearance, especially of the females, for the abdomens of parasitized flies were swollen with the mature female nematodes. The direct economic aspect of the case of parasitism reported herein lies in the fact that the maggots which feed upon plant roots are destroyed or rendered less active, and the fact that those fortunate enough to transform to the adult fly stage are rendered incapable of perpetuating their kind.

#### LIFE HISTORY OF TETRADONEMA PLICANS COBB

This nematode has but the one host, which may be larvae, pupa or adult of the *Sciara* fly. Possibly it may live also in other species than the *Sciara coprophila* in which it was found.

There is considerable dimorphism in the mature nematode. The females are large, reaching a length of 5 mm. or more, while the males are less than 1 mm. long. The striking characteristic of the female is shown in the mature egg laden form (Fig. 4). The swollen portion is caused by the storage of several thousand eggs beneath the cuticula, which serves as a retaining capsule. The notch on the ventral side of the worm marks the position of the genital opening. The relative sizes of the two sexes some time after mating is illustrated in Figure 3. Here the female has begun to lay her eggs as shown by the spindle-



shaped capsule; this continues to enlarge until the female has the appearance shown before but, barring accident, the eggs are not discharged until the death of the worm.

*Method of Infection.*—The nematode eggs very often have been induced to hatch in the presence of moisture. All efforts to observe the young larvae enter the maggot have failed. When first discovered in the host, they appear identical with those just escaped from the egg and have been detected first in the caudal end of the maggot in the region between the body wall and the digestive tract. The older maggots are much less susceptible to infestation than the younger ones. When young maggots not more than 3 mm. long were placed in earth containing the nematode eggs, they would, in the course of a day or two, be found to contain sometimes as many as twenty-one young parasites. The parasite probably gains access to its host through the alimentary canal. Even newly hatched maggots are of sufficient size to consume solid objects larger than the egg of the nematode. The larger maggots habitually swallow bits of earth and solid pieces of organic matter many times the size of the eggs. As a matter of fact, I have found the eggs of this nematode in the digestive tract of small *Sciara* larvae, and believe that the young nemas hatching from eggs that have been swallowed, bore through the wall of the digestive tract into the body cavity of the maggot.

*Percentage of Infection.*—As stated elsewhere in this paper, only one lot of infected material has been found. In this case every maggot was infected. Material from this lot was used to make artificial infections.

*Development and Behavior of the Host.*—The life cycle of this nematode has been followed from its earliest observed stage to the mating of the sexes and the formation of the egg capsules about the females, all of which takes place within the body of the maggot, and is clearly visible due to the transparency of infested larvae. The young nematodes as they are found in the maggot are of two kinds and of the same size as those that have hatched from eggs in a drop of water. This precludes any possibility of an alternate host. The method of studying the life cycle was to examine carefully young maggots taken from noninfested stock and then place them in small stenders with earth containing nematode eggs. Examinations were made several times daily. The eggs mixed with the debris swallowed by the maggots were not to be noticed in the living maggots, but when the latter were killed and the digestive tracts removed and cleared in cedar oil, as many as a dozen nematode eggs were found in the mid gut of one maggot. In the living maggots the first newly hatched

worms were to be noted in the body cavity at the posterior end of the body. Here they would often get into the way of the blood stream entering the dorsal vessel, and be buffeted back and forth. The slenderer worms were usually coiled at both ends, while the shorter plumper forms remained outstretched or slightly curved. From this time on the growth of the worms is rapid and usually timed, so that their eggs are produced before the maggot is ready to pupate. In cases of slight or late infection, the nematode cycle may not be completed until the pupal stage has been reached or even the adult fly produced. In adult flies have been found gravid females, heavily egg laden and also small nematodes equaling in size those of a few days of age. These I take it have been arrested in their development by the growth and maturity of older worms. Indeed even in a given maggot isolated for study the range of development of the various worms has been so great that I have found it difficult to tabulate the stages.



*a*, Caudal end of mature female. *b*, Female with two males attached.

On the average, very young parasites have been found in maggots from 1.5 mm. to 4 mm. long. Mating begins when the female nematodes are less than 1 mm. long, a size they may attain in from five to ten days. The remaining growth of the female, which attains some 5 to 7 mm. in length, and of the egg capsule involves the next two weeks.

*Mating.*—As the worms approach sexual maturity, marked activity is noted which may precede mating by a day or two. They begin to coil and uncoil about each other in an energetic manner. The males are the more active, coiling about the body of the female with the caudal end almost indiscriminately, until finally the genital opening is grasped, and the male rests with caudal end tightly clasped about the female, and the anterior end directed away at about a right angle. The spicules hold the males so firmly in position that they are often difficult to dislodge with dissecting needles. As many as three or four males may become attached to one female, and remain until the female completes

her egg capsule and dies. The males that remain unmated finally are relegated to the caudal end of the female, where they become granular in appearance and sluggish in action, finally dying. The males remaining attached to the female also show this granular appearance.

*Oviposition.*—The eggs of this nematode are retained beneath the thin cuticula of the female. As the female comes to the age of oviposition, the ova may be seen within her body in various stages of development. They appear both cephalad and caudad of the genital pore. Oviposition may be slow at first, and the first eggs can be seen to pass out and slip along under the cuticula. This is seen plainly in living females at the beginning of oviposition. The nature of this cuticular egg storage chamber may be studied in glycerin jelly mounts of such females in toto or by means of sections. The egg (Fig. 5) possesses a fairly thick shell, somewhat testaceous in color. It appears somewhat disk-shaped. The photograph shows them in flat view. When an egg is placed upon edge it forms an oblong outline, well shown in sections. It measures  $33.2\mu$  in diameter and  $16.6\mu$  in thickness. In the egg-burdened female will be found eggs in all stages of development, from unsegmented eggs to those containing actively coiling embryos, the latter not being abundant until the egg capsule is fairly well started.

NUMBER OF EGGS PER FEMALE PARASITE

Host	Size of Host	No. Parasites		Size of Parasites, Female	Total Number Eggs	Remarks
		Female	Male			
Pupa	Small	1	3	5 mm.	1,262	Embryos, all stages
Adult	Normal	1	6	5.2 mm.	2,046	Embryos, all but late coil stage
Larva	Enlarged	1	?	?	5,123	All stages of embryo
Larva	4 mm.	1	3	5 mm.	262	Egg capsule just beginning
Larva	6 mm.	1	2	4.9 mm.	1,763	Still egg laying
Larva	5 mm.	1	3	5 mm.	1,240	Still egg laying
Adult	Abdomen enlarged	1?	2	5.1 mm.	2,484	
Larva	5 mm.	1	1	5.1 mm.	5,520	
Larva	6 mm.	1	3	5 mm.	2,005	
Larva	7 mm.	1	2?	5 mm.	3,750	
Larva	7 mm.	2	5	5 mm.	4,700	

*Incubation and Hatching.*—Eggs containing mature embryos from young females, when dissected out and placed in water, hatch within twenty-four hours; eggs from old females hatch very shortly, from a few minutes to a few hours. The embryo may be seen coiling about within the shell for some time before it forces its way out. Newly hatched larvae are of two kinds, a very slender form with a curve at the caudal end  $125\mu$  long, and a plumper, slightly curved form  $90\mu$  long. The former, in hatching, often has some difficulty in freeing itself from the egg. In one case the little worm struggled for a half hour; it was free save for the caudal end, which hooked firmly into the

egg shell. The nema twisted about through the water, stopping now and then to give the whole body a series of vibrations in an attempt to get free. It was thirty minutes before a sudden and forceful effort set it at liberty.

*Dispersal.*—The distribution of this nematode may take place by migration of the maggots, through infested flies, and through the agency of air or water. The heavily infested maggot disintegrates and the nematode eggs mix with the soil to be eaten by other maggots or tossed about by wind or water. Dispersal by adult flies was proved in the laboratory. Three potted plants containing eggs and very young sciara maggots were placed in the same rearing cage with a can of earth containing infected maggots. From this can infected adult flies flew to the other pots, where some of them died. The dead bodies were full of nematode eggs which shortly brought about an infection of the maggots in the flower pots.

#### EXPLANATION OF PLATE

Fig. 1. Normal sciara maggot showing large white fat bodies and segmentally arranged discs of adipose tissue.

Fig. 2. Sciara maggot in advanced state of parasitism; white female nematode within.

Fig. 3. Mating female *Tetradonema plicans*; two males attached; egg laying already begun.

Fig. 4. Gravid female nematode swollen by eggs retained beneath cuticula.

Fig. 5. Eggs in various stages of development.

Fig. 6. Female nematode with reproductive organs dissected out.







## THE LONGEVITY OF THE FISH TAPEWORM OF MAN, *DIBOTHRIOCEPHALUS LATUS*

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While many species of tapeworms are known to be but short-lived within their host, it is well known that under favorable conditions others may live for a considerable period. This seems especially true of *Taenia saginata*, of which Leuckart says:

"The life of the present species seems to be very long. At any rate it is not at all rare for patients to evacuate proglottides almost daily for years. One of my Russian students harbored two tapeworms for more than five years. In another case the disease continued for more than eight years. Wawruch mentions several cases which lasted from twenty to twenty-five years, and in one case speaks even of thirty-five years."

There are also on record a number of cases of persisting infestation on man by the fish tapeworm, *Dibothriocephalus latus*, some of them evidently unquestionable, others complicated by the possibility of reinfection. Two cases in the clinical records of the University Hospital, Minneapolis, Minnesota, seem especially clear cut and of such interest as to justify publication. For permission to use the data I am under obligations to Dean E. P. Lyon of the Medical School.

1. Mrs. X—, for four years a resident of Minneapolis, was admitted to the hospital July 12, where on August 24, she gave birth to a child. On August 31 it was noted that her stools contained segments of *D. latus* and ova of the same. September 1, after appropriate treatment, she evacuated a complete specimen, with head, of *D. latus*.

The patient was a Russian Jew who had left Russia some five years previously. She had been "troubled a good deal with gas on the bowels and cramps during the past five years."

2. Mrs. Y—, a Swedish woman, 43 years of age, was admitted to the University Hospital October 13, 1911, with a complication of diseases. On account of a high degree of eosinophilia which the patient maintained while in the hospital the possibility of parasitic infestation was suspected, and on examination of the stools many eggs of *D. latus* were found. Treatment was instituted and a worm recovered complete, after which the eosinophilia dropped from 25 to 2.8 per cent.

The records show that tapeworm segments were again found in the stools on February 18, 1912, and March 4, but no heads were found. The patient was discharged, but was readmitted July 3, 1912, with diagnosis of "*B. latus*, Addison's disease, tuberculosis of the lymph glands." After treatment, 15 cm. of the worm, but no head, were discharged. Eggs and segments continued to be noted in stools up to August 21, when, following treatment, some 20 m. of worms and two heads were discharged.

The patient stated that she had had worms since fifteen years of age. From the time when she first began to menstruate at the age of 14, she often noticed worm segments, and sometimes was able to extract long pieces of worm. The segments were of a very white color. One day she told her mother about it, but as they never bothered her, she neither consulted a doctor nor at any time took any medicine to expel the worm.

She was never ill until she was 29 years of age. It was at this time that she came to the United States from Sweden, and while aboard ship she became very sick. She vomited severely, the vomitus containing pieces of the tapeworm. The worms from which these segments were derived had not been expelled previous to the patient's entering the hospital.

Since coming to the United States she had lived for two years in Brooklyn, three years in Michigan, three years in Wisconsin and five years in Minnesota.

Though it is clear that the first infestation in this case occurred at least twenty-nine years previously, this, of course, does not prove that the age of the worms expelled was that great. Under appropriate conditions of environment and food habits, repeated infections may have occurred during the early life of the patient. The age of the worms recovered at the hospital was at least the thirteen years covered by the period of residence in this country—how much greater, it would be impossible to judge.

Concerning both of the cases here reported, it may be objected that there is evidence that *Dibothriocephalus latus* is endemic in some sections of this country. That this does not account for these cases seems evident, from the extreme rarity of native infestation, the fact that both patients had lived in the large cities, rather than in the region of the suspected lakes, and especially from the clear history of infestation before coming to this country.



## WINTHROP DAVENPORT FOSTER

Winthrop D. Foster died in Washington, D. C., on October 6, 1918, as a result of pneumonia complications following an attack of influenza. He is survived by his wife, formerly Miss Christian Kershaw of Windsor, Ontario, and three children. Mr. Foster was born in Jersey City, N. J., December 28, 1880, and was the son of a Congregational clergyman, Dr. Addison P. Foster. The family was from New England, and Mr. Foster attended the Roxbury Latin and Newton High Schools, graduating from the latter in 1900. He later attended Williams College, from which he received the degree of B.A. in 1904, and the degree of M.A. in 1912.

He also studied forestry at the University of Michigan and veterinary medicine at George Washington University. In 1904-05 he was instructor in biology in Assumption High School at Assumption, Illinois, and from 1908 to 1910 was clerk-translator in the Census Bureau. In 1910 Mr. Foster became Junior Zoologist in the Zoological Division of the Bureau of Animal Industry, and was connected with that division up to the time of his death. He was one of the first members of the Helminthological Society of Washington and was also a member of the Biological Society of Washington.

Mr. Foster's work in parasitology was done principally on the parasites of swine and along the lines of critical tests of the efficacy of anthelmintics, though he published a number of notes on other lines.

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## NEW HUMAN PARASITE

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*Enteromonas bengalensis* Chatterjee, 1919. This flagellate was found in India in the stool of a ten year old boy who died after suffering a year from chronic dysentery. It differs from *Enteromonas hominis* Da Fonseca, 1915, in that the nucleus does not show a differentiated karyosome; also there is no blepharoplast attached to the nucleus by a rhizostyle. (Indian Jour. Med. Research, 6: 380-382, text fig., Jan., 1919.)

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## NOTES

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William Walter Cort (A.B., Colorado College, 1909; Ph.D., University of Illinois, 1914), who is at present on the staff of the University of California, and consulting helminthologist of the California State Board of Health, has been appointed Associate in Helminthology in the School of Hygiene and Public Health, Johns Hopkins University. His work in Baltimore will begin in the fall.

Ernest Carroll Faust (A.B., Oberlin College, 1912; Ph.D., University of Illinois, 1917), now instructor in Zoology at the University of Illinois, has accepted a position with the China Medical Board, Rockefeller Foundation, as Associate in Parasitology, Department of Pathology, Union Medical School, Peking, China. He plans to assume his duties in Peking early in October.

The Instituto Bacteriologico of Buenos Aires has been reorganized. Among the departments and appointments listed are Parasitology, Doctor Wolffhügel; Medical Zoology, Doctor Bachmann. The latter has been designated for a mission abroad.

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